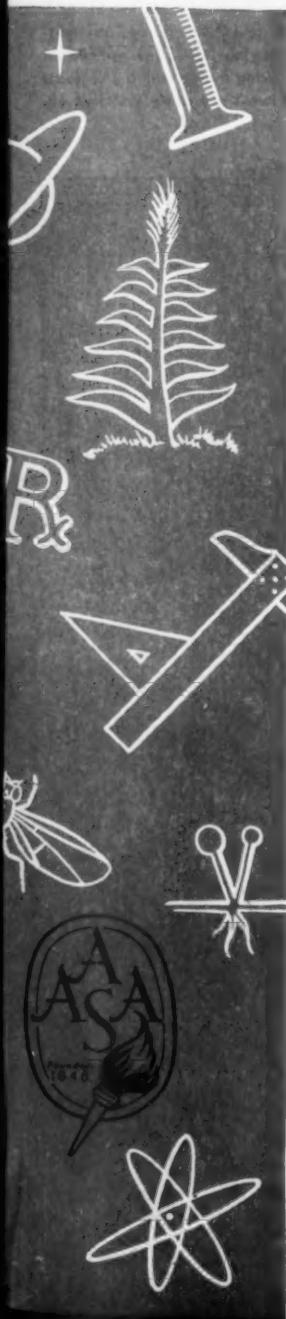




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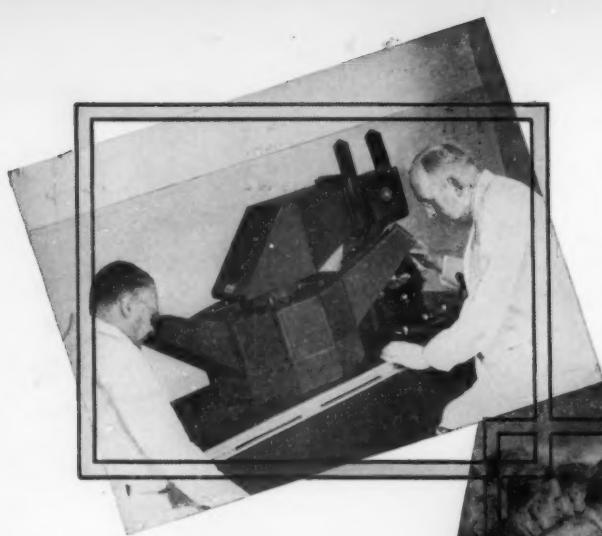
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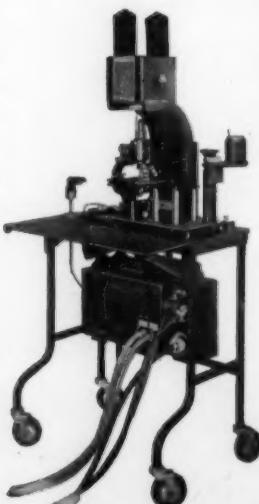
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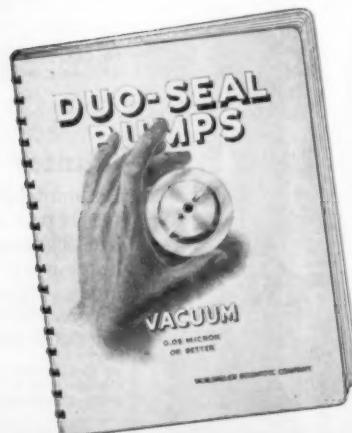
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Ecological Taxonomy

THE article "Taxonomy Today" by Robert Cushman Murphy (SCIENCE, 115, 3 [Feb. 1, 1952]) depicted the relatively strong position that taxonomy properly holds, because (a) only about 15 per cent of the fauna is known and more needs to be known; (b) a chaotic world of life is being replaced by one of order through taxonomy; (c) a large proportion of the known myriad physical manifestations of life are on permanent and accessible file in museums; and (d) the modern taxonomist regards his specimens as representing genetic complexes in protoplasm that display infinite varieties of patterns evolved under a condition that man cannot "control"—the amassed effect of secular time.

To this important array of values of taxonomy should be added others. Much time and energy have been devoted by taxonomists to isolating morphological patterns of species and subspecies and determining the geographic ranges of each. This is only a steppingstone to further progress in many lines—units around which accumulations of knowledge could be formed for comparison with one another. Until such units are stabilized so that they can be recognized, specific knowledge cannot accumulate—it will of necessity be generalized because, without such standardization, one worker cannot add to the specific knowledge of others.

Many species and subspecies occupy not only geographic ranges but also ecological ranges, delimited by certain types of habitats, certain types of biota, and certain niches within such habitats or biota. If these ecological ranges can be established, it will facilitate study of the divergent characters by means of which species and subspecies have been developed during long periods of time under the aegis of natural selection and under different conditions of habitat.

The ecological range can be determined in much the same way as the geographic range and by similar methods of specimen collection. The need for recording the habitat and biotic unit points to the necessity

for standardized ecological classification, with units that can be recognized and used in collecting such data. The ecological point of view is already infiltrating taxonomy but, if extended and incorporated, would lead to better concepts of species and subspecies relationships and would yield a better foundation for understanding the ecological units of the biota found in nature. If the ecological point of view is increasingly adopted in taxonomy, then taxonomists would be equipped to move into the field of ecology and establish the taxonomy of ecological units.

This would necessarily involve a shift in basic units, but similar rules of nomenclature could be applied. The basic units, which would, of necessity, deal with both plants and animals, should (1) be natural subdivisions of the total biota and not merely special groups of plants or of animals; (2) comprise groups of species that interact with one another and evolve together; (3) occupy geographic areas that may be either continuous or discontinuous; (4) be distinctive aggregations of species; (5) be aggregations of species that are more or less interdependent; (6) be groups occupying habitats that are more or less distinct from other habitats; (7) be groups that are more or less independent of other communities (minor exchanges); (8) be aggregations that have histories as communities and that may have pioneer, developmental, and stable stages; (9) have boundaries, but these may be relatively abrupt or intergrade with adjacent units; (10) have recognizable physiognomies that can usually be delineated in the field.

If a species be regarded as potentially an interbreeding population of similar individuals, then this ecological unit differs in being an ecological aggregation of interacting species that evolve to fit one another. This approach could lead to a taxonomy of ecological units, to a study of the mechanism involved in natural selection aspects of evolution, and even to an experimental taxonomy.

ANGUS M. WOODBURY

Department of Vertebrate Zoology
University of Utah

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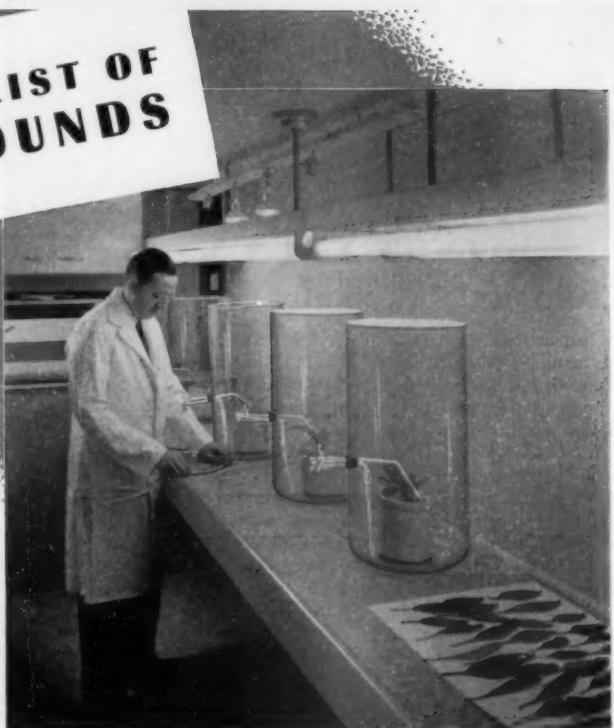
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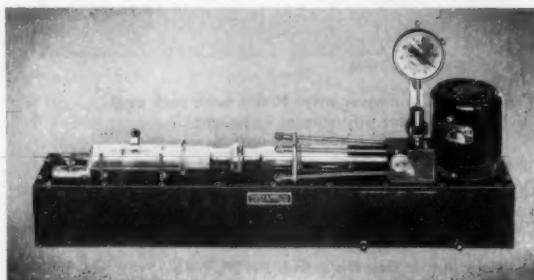
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NATIONAL ACADEMY OF SCIENCES

Abstracts of Papers Presented at the Annual Meeting April 28-30, 1952, Washington, D. C.

Utilization of Carbon Dioxide in the Synthesis of Protein and Nucleic Acid by *Escherichia coli*

E. T. Bolton and Philip H. Abelson
Carnegie Institution of Washington

Cells of *E. coli* take up carbon dioxide from the bicarbonate of a synthetic medium which contains glucose as an energy source. The carbon of the NaHCO_3 molecule is utilized in the synthesis of proteins and nucleic acids. When $\text{NaHC}^{14}\text{O}_3$ is used the isotope is found largely in aspartic and glutamic acids, arginine, lysine, proline, and threonine, but not in the remaining amino acids of the proteins. The location of the tracer isotope within the amino acid molecule is characteristic of the amino acid and does not change for a variety of altered culture conditions. Both the purines and pyrimidines of the nucleic acids become labeled with C^{14} after growth of cells in $\text{NaHC}^{14}\text{O}_3$ media.

When the culture media are supplemented with non-radioactive metabolites such as amino acids or purine bases, the labeling of certain amino acids or nucleic acid constituents, which would occur in the absence of supplementation, is specifically suppressed. It is ordinarily observed that a supplemental compound—e.g., an amino acid—is utilized by the cell for the construction of protein in preference to synthesis *de novo* of the same compound from the simpler constituents of the medium. Metabolic intermediates are also preferentially utilized. For example, citrulline will completely eliminate the utilization of CO_2 for arginine synthesis. These observations have provided a basis for the determination of some biosynthetic interrelationships among a number of the amino acids of the proteins, and among the purines and pyrimidines of the nucleic acids of *E. coli*.

These studies have shown that there are at least five distinct processes by which carbon dioxide is assimilated by *E. coli*: those giving rise to (1) the purines, (2) the pyrimidines, (3) aspartic acid, (4) glutamic acid, and (5) the amidine carbon of arginine. It is significant that these pathways for carbon dioxide utilization are essentially identical with those that have been individually discovered in a number of different living systems.

The Role of Glutathione in Protein Synthesis by *Escherichia coli*

R. B. Roberts and E. T. Bolton
Carnegie Institution of Washington

One fourth of the total sulfur of *E. coli* cells can be extracted with cold trichloroacetic acid. This soluble fraction contains one major sulfur component identified as glutathione (GSH). The biosynthesis of S^{35} -labeled GSH from $\text{S}^{35}\text{O}_4^{2-}$ is blocked by the presence of unlabeled cysteine or GSH in the medium but not by the presence of unlabeled methionine or homocystine. Kinetic studies show that GSH has a high rate of turnover.

When cells containing S^{35} -labeled GSH grow in the presence of $\text{S}^{35}\text{O}_4^{2-}$, roughly one half the S^{35} of the GSH is incorporated into the proteins and one half is released into the medium. With methionine added to the medium

the transfer of GSH³⁵ sulfur to protein-methionine is blocked; with cysteine the transfer to both the protein-cysteine and the protein-methionine is blocked. The sulfur released into the medium can be separated into several fractions by ion exchange resins or by paper chromatography. These fractions contain GSH and several other organic S-compounds including peptides.

C^{14} -labeled GSH is synthesized by cells growing in a medium containing C^{14}O_2 , mineral salts, and glucose, but this synthesis is blocked by the presence of unlabeled glutamic acid or GSH in the medium. GSH also suppresses the incorporation of C^{14}O_2 into several amino acids of the proteins.

These findings demonstrate the important role of glutathione as an intermediate in the synthesis of proteins by *E. coli*. Some progress has also been made in determining the mechanism of utilization of glutathione.

Amino Acid Sequence in Proteins

Philip H. Abelson, *Carnegie Institution of Washington*

The number of individual proteins to be found in the living world is enormous. This diversity arises largely from combinations of approximately 20 amino acids used as building blocks. The demonstration of recurring patterns in the order of amino acids in the proteins would greatly simplify our understanding of protein synthesis and structure. Examination of the products of partial hydrolysis of the total proteins of *E. coli* has uncovered one such pattern.

Attention was focused on the combinations of amino acids with cysteine. For these studies the cysteine of *E. coli* was specifically labeled with S^{35} by growth of cells in a synthetic medium containing $\text{S}^{35}\text{O}_4^{2-}$ and non-radioactive methionine. The cells were harvested, freed of nonprotein constituents, treated with performic acid, and partially hydrolyzed to obtain a mixture consisting principally of dipeptides, tripeptides, and tetrapeptides. A combination of electrophoresis, resin chromatography, two-dimensional paper chromatography, and radio-autography facilitated purification and isolation of peptides containing cysteine acid.

More than 75% of the cysteine acid was found in amino acid combinations which included glycine. A peptide containing only cysteine acid and glycine in that order accounted for more than 10% of the original cysteine of the proteins of the bacterial cells. On the basis of a purely random order of amino acids in proteins a maximum of 4% of the cysteine could have been present as cysteinyll-glycine.

The presence of the cysteinyll-glycine combination in glutathione and the wide occurrence of the latter throughout nature are suggestive of a possible widespread occurrence of the cysteinyll-glycine combination in proteins. Indeed, Sanger and Tuppy have recovered cysteinyll-glycine from beef insulin following performic acid treatment and partial hydrolysis. Their fraction "B" of insulin was shown to possess two cysteine molecules, both followed by glycine.

Other possible regularities in patterns of amino acid sequences are under investigation.

Effects of Testosterone Propionate on Social Status in Six Breeds of Common Domestic Hens

W. C. Allee and Darhl Foreman

University of Florida and University of Chicago

This study in comparative sociology has focused on results of daily injections of male hormone into hens of low social rank in small flocks of the following breeds: Light Brahma (Asiatic); Barred Plymouth Rock, Rhode Island Red, New Hampshire (American); Ancona, White Leghorn (Mediterranean); and the selected single-comb progeny of a cross between White Leghorns and White Wyandotte (American), frequently called the "English" strain of White Leghorns.

Androgen-injected hens became more aggressive in all flocks except in "English" Leghorns. Sixty-four per cent of treated individuals gained higher social status; one of them reached highest social rank among Anconas, uncrossed Leghorns, Plymouth Rocks, and Brahmams. Considering all injected birds in each breed, Anconas were clearly most responsive; Leghorns also showed good reactivity, indicating high effectiveness of the androgen on these light-weight, excitable Mediterranean breeds. Despite the small numbers tested, refractory individuals were found in all breeds tested, except Ancona.

The American breeds, as a group, gave less response to androgen treatment than the Mediterraneans and, Plymouth Rock excepted, were no more responsive than the heavy phlegmatic Brahma. Not counting "English" White Leghorn as a breed, the difference in response between the two White Leghorn strains was greater than that between any two breeds tested.

The characteristic differences in peck-order relations in these breeds are, to some extent, androgen-induced. The interbreed variations may be in part, though not wholly, a matter of dosage in relation to weight. We do not know the mediation processes whereby male hormone influences the fundamental breed differences in behavior.

Poliomyelitis and the Weather

Charles Armstrong, National Institutes of Health

A new concept is suggested for explaining the peculiar seasonal incidence of poliomyelitis, which views the upper respiratory tract of man as either the portal of entry or of exit for that portion of virus largely effective in its transmission.

When atmospheric air is inhaled it is warmed in the nose and throat to 90° F and its relative humidity raised to 90%. The increased secretions, and possibly other alterations, necessary to prevent the mucous membranes from becoming parched in cold weather are believed to hinder the transmission of poliomyelitis, whereas a quiescent membrane with a minimum of secretion and evaporation, such as prevails in warm moist weather, favors it. It is felt that the condition of the mucous membranes of the nose and throat is of more significance for the spread of poliomyelitis than is the mere opportunity for the virus to spread from person to person.

This concept relates the summer incidence to a recognized alteration in the upper respiratory passages, rationalizes the long-recognized correlation of poliomyelitis incidence with warm weather, and presupposes a

similar and possibly more significant correlation with humidity, not heretofore apparent. However, when inspired air warmed in the nose and throat to 90° F is considered, a direct correlation between atmospheric relative humidity at this temperature and the incidence of poliomyelitis becomes apparent. The curves where so far studied are so similar in general and in detail as to indicate a significant relationship.

Genic Control of Gene Expression in *Paramecium aurelia*

Geoffrey Beale, University of Edinburgh

The hereditary properties of an organism depend, as is well known, on its genes, but these latter can only function satisfactorily in certain environments. Further, the cytoplasm is known, from work with *P. aurelia*, to control which of a series of genes will be fully expressed and which inhibited. Consequently, the direction in which a given cell will develop depends on the state of the cytoplasm. Evidence will be presented showing that this cytoplasmic state is partly controlled by the external environment and partly by the genes themselves.

In variety 1 of *P. aurelia* there is a series of genes—*d*, *g*, *s*—controlling the formation of antigenic substances on the cilia. The *d* alleles are expressed in a cytoplasmic state normally formed at a high temperature (29°–33°), the *g* and *s* alleles at medium (25°) and low (18°) temperatures, respectively.

The readiness with which the cytoplasm changes from one state to another under the influence of changing temperature, is markedly different in different races of the organism. The genetic factors controlling these diversities have been investigated by a series of matings, including repeated backcrosses by which the antigen-determining genes of one race were combined with a background of other genes of another race. It was found that the stability of cytoplasmic states depended partly on the antigen-determining genes themselves and partly on other genes.

Hence there is a mutual interaction between genes and cytoplasm whereby the cytoplasm determines whether a given gene shall be expressed, yet is itself partly controlled by the same gene.

The Limiting Negative Pressure of Mercury in Pyrex Glass

Lyman J. Briggs, National Bureau of Standards

It is generally considered that mercury does not "wet" glass, as evidenced by the convex surface of the mercury meniscus in a barometer. If this were true under all conditions, the development of a negative pressure in a mercury column would be impossible. But pure mercury in a clean evacuated degassed tube shows a flat meniscus. In other words, when the tube is degassed, mercury does adhere to the glass wall.

A U-tube manometer of 5-mm bore with one leg sealed off was evacuated and degassed at 500° C with a mercury-vapor pump, and then filled with pure mercury by distillation. The vertical height of the mercury column from the top of the closed leg to the meniscus in the open tube was 52 cm. When again evacuated, the mercury column remained suspended from the top of the tube. In other words, the column showed a negative pressure of two thirds of an atmosphere. But when once released by vertical jarring, the column would not hang up again.

In endeavoring to extend these measurements, short mercurial thermometers were first used. Each thermometer was cemented to the spinner of a variable-speed motor, with the spin axis bisecting the distance from the end of the bulb to the meniscus. The maximum stress on the free capillary column was consequently at the spin axis. The results for nine thermometers were erratic, giving negative pressures ranging from 2 to 17 bars.

Finally, freshly-drawn capillary tubes with fine bores were evacuated, degassed at about 550° C, and filled by breaking one end under pure mercury. This procedure gave limiting negative pressures of mercury in Pyrex glass of 46, 47, and 58 bars at room temperature, as compared with about 10 bars in a similar ungassed capillary. Degassing thus appears to result in a fourfold increase in the adhesion of mercury to glass.

The Cytogenetics of Introgession

Roy E. Clausen, *University of California, Berkeley*

The cytogenetical problem of introgression, as exemplified in transfer of genes from wild species of *Nicotiana* to cultivated tobacco, *Nicotiana tabacum*, involves special features owing to the high sterility of the F_1 hybrids and to the low degree of association of chromosomes of the two species in the hybrids. Thus in the transfer of necrotic mosaic resistance from *glutinosa* to *tabacum*, the normal hybrid is almost completely sterile and the chromosomes of the two species exhibit only a low degree of association. The sterility may, however, be overcome by crossing tetraploid instead of diploid *tabacum* with diploid *glutinosa*, producing in the first instance a relatively fertile triploid hybrid having two sets of *tabacum* and a single set of *glutinosa* chromosomes. Such a hybrid, either when selfed or when backcrossed to *tabacum*, quickly eliminates the alien *glutinosa* chromosomes; but if selection is practiced for a specific feature, such as necrotic mosaic resistance, a $24_{11} + 1_1$ segregant is first obtained, which, in addition to the complete diploid set of *tabacum* chromosomes, retains the single alien chromosome carrying the selected factor. Introgression of the alien gene into the *tabacum* complex next ensues as a consequence of association of this extra alien chromosome occasionally with a specific *tabacum* chromosome, where, by following simple segmental interchange, a whole alien chromosome may be substituted for a specific *tabacum* chromosome (alien substitution) or the segmental interchange chromosome may be substituted for it (segmental substitution). The shift of the gene from the extra chromosomal condition either to alien substitution or to segmental substitution is signalized by appropriate alterations in the transmission ratio of the selected character.

Nuclear Cytology of the Fungus *Eremascus albus*

Edward DeLamater, Sidney Yaverbaum, and Lucille Schwartz, *University of Pennsylvania*

Current evidence is conflicting concerning the nature and position of the nucleus in the cells of the lower fungi, including the yeasts, the number of chromosomes, and their characteristics. The mechanism of their division is likewise not clear. Since the advancement of the genetics of the lower fungi depends largely upon a definition of the nucleus, pertinent evidence is important.

The vegetative nuclear divisions in *E. albus*, Eidam, proceed by mitosis. The nucleus is distinct from other cell inclusions, including central vacuoles and basophilic granules.

Sexuality occurs as a fusion between two like gametangia to form a single sexual cell in which the gamete nuclei fuse. In the sexual cell the differentiation of the large central vacuole and the nucleus is most distinct.

In the fusion nucleus the chromosomes occur as long, tangled threads in a characteristic leptotene stage. In the zygote the chromosomes pair, at first at one or two points along their lengths. Subsequently they pair along their entire lengths, and enter pachytene. The chromosomes contract in the classic manner through typical diakinesis. At metaphase, six pairs of chromosomes are visualized. The centrioles and spindle appear to have an extra nuclear origin. As anaphase and telophase proceed, no new nuclear membranes are formed about the daughter nuclei. Meiosis II follows directly. Only following the third division are nuclear membranes redelineated and spore walls laid down.

The observations presented constitute: (1) the first clear demonstration of the nucleus and its *modus operandi* in this group of organisms; (2) a characterization of its chromosomes; and (3) a demonstration of a remarkably classic pattern of nuclear structure and activity for these organisms.

On Finite Groups with Two Independent Generators, II

Jesse Douglas, *Columbia University*

This paper continues with the developments begun in a previous paper of the same title, presented to the Academy in November 1951. Necessary and sufficient conditions governing the defining substitutions θ, φ of a two-generator group $\Gamma = \{A^{\pm}B^{\pm}\}$ (conjugate special substitutions) are given. Included is the simple condition: $\theta(0) = 0, \varphi(0) = 0$. Each substitution θ (or φ) of such a conjugate pair has by itself some very special properties. Characteristic is the following: every 0-fixing translation of θ must be a power of θ . "Translation" means a transformation $x = x' + a, y = y' + b$, converting the substitution $x \rightarrow y$ into $x' \rightarrow y'$. An equivalent condition is: the 1-translation of any power θ^n of θ must be again a power of $\theta: \theta^n 1^{(s)}$. This leads to the idea of derivatives θ_s of any special substitution θ . If $\theta, \theta_1, \theta_2, \dots, \theta_k, \dots$ denote the series of successive derivatives, the type of θ is defined as the least index k such that $\theta_k = (0)$ —the identity on the range consisting of the single element 0. Examples of substitutions of various types, up to 6, are given.

Plaques Produced on a Monolayer Tissue Culture by Single Particles of an Animal Virus

R. Dulbecco, *California Institute of Technology*

Plaque formation by single virus particles is the basis for the analytical techniques used in the study of bacterial viruses. This has now been obtained with an animal virus, the virus of Western equine encephalomyelitis growing on chicken fibroblasts. This phenomenon may serve for the development of an animal virus assay much superior to any other assay now in use, and for the isolation of the progeny of single virus particles.

Virus plaques are produced on a continuous fibroblastic layer grown on the bottom of a glass flask of the size of a Petri dish. A continuous cell layer is first grown in two days, following a technique devised by W. R. Earle, and then the virus is applied on it; easily detectable round necrotic areas are produced within two or three days of subsequent incubation.

The number of plaques produced by two different dilutions of the same virus sample is proportional to the concentration of the virus; hence a plaque is produced by a single virus particle. The comparison between the number of plaques produced and the fraction of chicken embryos infected by the same virus sample indicates that all or nearly all virus particles able to infect the embryo produce a plaque. There is therefore a 1:1 relation between infecting particles and plaques.

Plaque production is technically simple and reproducible.

Experiments are under way in which this phenomenon is applied to a detailed analysis of this virus-host system.

On the Dynamics of the Cochlea and the Middle Ear

Harvey Fletcher, *Columbia University*

The fundamental differential equations of force and continuity for the liquid in the cochlea are derived. Coupled to the motion of this liquid is the reaction of the basilar membrane separating the two canals of the inner ear, the oval window membrane, the round window membrane, and the helicotrema. These reactions are taken into account and an equation derived which gives the velocity and displacement of the basilar membrane at any position in terms of the pressure level and frequency of a tone impressed upon the eardrum. Calculations using this equation show that the position of maximum stimulation of the nerve endings along the basilar membrane is near the stapes end for the high frequencies, and near the helicotrema for the low frequencies.

By making the rational assumption that the number of nerve discharges originating from any small part of the basilar membrane is proportional to the square of the velocity of the membrane at that position, it is shown how to calculate the sound pressure levels corresponding to the threshold of hearing at various frequencies. These calculated values agree with those observed. This calculation shows that the poor acuity of the normal ear for perceiving low-frequency sounds is due primarily to the poor transformer actions of the middle ear.

Uterine Influences upon Intrarenal Blood Distribution

Kenneth J. Franklin, *University of London*

The following résumé is based upon the literature, public statements, and experimental results obtained alone or with colleagues.

In specific cases of renal trouble (oliguria, anuria, acute tubular nephrosis, bilateral cortical necrosis) in pregnant and puerperal women there has been evidence of excessive uterine distension by products of conception / concealed accidental haemorrhage / cortical ischaemia with medullary flow continuing / excessive uterine contraction *post partum*.

Diversion of the renal cortical blood flow can occur physiologically in nonanesthetized animals, and spontaneously in anesthetized animals of various species.

In rabbits it has been found that:

1) Distension of either horn of the virgin, parous, or pregnant uterus with Ringer's solution, the animal's blood, or blood from a donor animal can cause diversion of blood flow from the cortices of both kidneys (a) by direct reflex action upon the renal vasomotor apparatus,

(b) by a reflex *cum* humoral action dependent upon the integrity of the adrenal glands.

2) During spontaneous parturition there is a humoral (probably posterior pituitary) reduction of the renal cortical blood volume.

3) For a while after such parturition there is reduction of the same blood volume through uterorenal reflex action.

4) Appropriate stimulation of the renal nerves has caused acute tubular nephrosis and renal cortical necrosis.

Chain Polymerization of Bi-Bolaform Electrolytes

Raymond Fuoss and Paul Goldberg, *Yale University*

Ordinary 1:1 electrolytes show a moderate degree of association in methanol, where the electrostatic potential energy of a pair of ions at contact is of the order of several *kT*. Bolaform electrolytes are multivalent electrolytes in which one ion has the structure (+) . . . (+) or (-) . . . (-). If we consider a bi-bolaform electrolyte such as $\text{Me}_2\text{N}^+ (\text{CH}_2)_5^-\text{NMe}_2 \cdot \text{O}_2\text{C}(\text{CH}_2)_4\text{CO}_2^-$ in a solvent where electrostatic forces are sufficiently strong, we might expect chain formation of the sort schematically represented by (- . . . -) [(+ . . . +) (- . . . -)]* (+ . . . +). Experimentally such association should be detectable as an abnormally large viscosity. Our results demonstrate the expected effect: the reduced viscosity of the above salt in anhydrous methanol is 0.045 (i.e., several times the Einstein limit for spheres). The shape of the curve is similar to that for neutral polymers and in no way resembles the square root curve of the Debye-Falkenhagen theory for strong electrolytes. The following variables have been studied: dielectric constant of solvent, spacing of charges in the bolaform electrolyte, and concentration of added simple electrolytes. These compounds represent a new type of synthetic chain molecule which shows both characteristic similarities and contrasts to polyelectrolytes and polymers.

Homoeotic Mutants and Evolution

Richard B. Goldschmidt, *University of California*

I have claimed repeatedly that homoeotic mutants, as found in *Drosophila*, are good models of macroevolution by large steps (saltations). A new type of tetralterism is apt to illustrate how a single mutant affecting early embryonic processes can have effects of a macroevolutionary order. The main feature is that the localized action upon a specific developmental process may lead to a set of embryonic regulations and integrations that result in a widely divergent architecture of a part of the body. In the example, a mutant of the *podoptera* group, the embryonic separation of the anterior wing field from the rest leads to (1) reconstitution of this part into a leg or haltere-like structure (depending upon the size of the wing segment involved) by production of a mirror image half, (2) redetermination of the rest of the wing anlage into thorax, which is developed in the lower grade of the effect as a winglike appendage carrying the thoracic bristles; in the higher grades, corresponding to earlier action, the extra half-thorax is completely integrated with the normal part of the thorax into a new type of aberrant thorax, which, nevertheless, is perfect as such. The existence of all transitions proves the interpretation. The problem of the establishment of such a macromutant in nature will be touched upon.

Evaporation from Liquid Surfaces in Vacuum

K. D. Hickman and D. Trevoy, *Eastman Kodak Company*

The accommodation coefficients of liquids at the interface with vapor or foreign gas have been the subject of wide study in many laboratories under the tacit assumption that the coefficient with the pure saturated vapor will be characteristic for each type of liquid and generally less than unity. Experiments to be reported suggest that the coefficient varies over a tremendous range for any liquid according to the momentary and local circumstances at the surface, and is not dependent on chemical composition except under rigidly defined circumstances of purity and surface demand. It is found that the accommodation coefficient decreases when equilibrium with the vapor is disturbed and certain kinds of foreign molecules are present, and it is thus lowest when an impure fluid is evaporating freely into a high vacuum. Relative ratio of emission of vapor is now referred to as the evaporation coefficient; and it can be shown that there are definite rules governing the emission, the coefficient decreasing when the surface is pushed, increasing when it is stretched, and the values are repeatable and thus predictable with an accuracy of a few per cent. The low coefficients appear to be due to a traffic jam at the surface caused by molecules of impurity becoming stranded in the surface mosaic, where they limit egress of the solvent molecules. The surface skin has high mechanical strength, but it vanishes as soon as the causative conditions are removed.

The apparatus that led to these conclusions consists of a pot still and a sensitive probe which records the emission of vapor from a chosen point on the surface from moment to moment. The still contains means for stretching, pressing, or agitating the surface in known degree, and it is attached to a purification train which can, at will, continuously distill and overflow the contents. Only when the surface is purer than (it is believed) has ever been obtained before does the coefficient rise to unity, and then the value is not altered by pushing, pulling, or stirring.

A Coordinate System for the Treatment of the Motion of a Shell

R. H. Kent, *Aberdeen Proving Ground*

In dealing with angular motion of a shell about its center of gravity, it is convenient to use a spinning non-Galilean coordinate system. Two of the most widely applied treatments, using such systems, are those by Fowler, Gallop, Lock, and Richmond and by Nielsen and Syng, especially as extended by Kelley and McShane. The former group uses a coordinate system which turns with the particle trajectory. In this they consider two vectors, Λ , a unit vector representing the direction to the axis of the shell and, X , a unit vector having the direction of the actual trajectory. Nielsen and Syng use a coordinate system associated with the shell. They have three unit vectors, i_1 , i_2 , i_3 . Of these, i_1 has the direction of the axis of the projectile, and the other two are perpendicular to i_1 . The author proposes a system which differs slightly but significantly from that of Fowler, Gallop, Lock, and Richmond. In his treatment, the unit vector Λ is defined not with respect to a coordinate system turning not with the particle trajectory but with the actual trajectory. In this way the treatment of the vector X is avoided.

The method of deriving the equation for the complex

yaw is briefly explained. The results are compared with those of Fowler, Gallop, Lock, and Richmond, and of Kelley and McShane.

Sign and Symbol in Bee Communications

A. L. Krober, *Columbia University*

Von Frisch's searching experiments on the so-called language or communication of bees prove that these social insects transmit to one another, by dance-like motions, information as to the abundance, distance, and direction of supply of food. Surprisingly, all three of these kinds of information are quantitative. The communication of the first two sets of facts, on abundance and distance, could be explained as quantitative reflex responses to stimuli, which would expectably be genetic and congenital. However, the expression of direction, when a simple pointing repetition is not feasible, is by a movement the axis of which departs from an antigravitational vertical by an angle corresponding to the angle by which the direction of food supply from the hive departs from the direction of the sun from the hive. The relation of this means of expression to the fact denoted seems indirect, physiologically arbitrary, and abstract. In short, the expression resembles a linguistic expression, a true symbol, and not a direct sign or signal, although it is generally accepted that the faculty of symbolization and abstraction is restricted to human beings and is perhaps their most significant differentiating characteristic. The possibility or probability of interpretations contrary to the above is examined; but what analysis makes clearest is the importance of further directed observations and experiments on the problem.

Pure Quadrupole Spectra of Solid Chlorine Compounds

Ralph Livingston, *Oak Ridge National Laboratory*

The pure quadrupole spectra of a number of covalently bonded chlorine compounds have been observed as solids at low temperatures. A precise measure of the nuclear quadrupole moment ratio of the two stable chlorine isotopes is obtained from the determined quadrupole couplings. A comparison of quadrupole couplings gives a measure of the variation from one molecule to another of the electric field gradient experienced by the chlorine nuclei. The electric field gradient has its origin primarily in properties of the chemical bond that holds the chlorine in the molecule. It is found that the magnitude of the quadrupole couplings increases smoothly in the sequence CH_2Cl , CH_2Cl_2 , CHCl_3 , and CCl_4 , as well as in the sequence tert-butyl chloride, isopropyl chloride, and ethyl chloride. In these cases the changes in coupling may be explained by changes in ionic character in the carbon chlorine bond. A large number of other molecules, including many "Freons," correlate well.

The Electron Affinity of Iodide in Liquid Ammonia and the Heat Content of the Electron in Ammonia and Water

W. M. Latimer, R. E. Connick, and W. L. Jolly
University of California, Berkeley

The thermodynamic data are available for the calculation of the ΔH of the reaction in liquid ammonia.

$$I_{(am)} = I_{(am)} + e_{(am)}$$

$$\Delta H = 109.3 \text{ kcal}$$

The absorption curve for iodide in liquid ammonia has

been measured, and the maximum lies at 2540 Å. This corresponds to an energy of 112.4 kcal. If the photo-absorption results in complete ionization, the difference between the two energies should represent the relaxation energy of the solvent molecules about the iodide ion after the emission of the electron.

It has been generally assumed that the relaxation energy should be of the order of magnitude of the solvation energy. For iodide in ammonia this should be about 80 kcal. We are unable to give a completely satisfactory explanation for the small value, 3.1 kcal, calculated from the above data.

A comparison of the energies for the photodissociations of I^- , Br^- , and OH^- in water with the thermodynamic data indicates that the relaxation energies in water also are small. If it is assumed to be 4% of the hydration energy, as found for I^- in ammonia, one then calculates for these cases a consistent value of 89 kcal for the heat content of the electron in water.

Aurora Borealis

Donald H. Menzel, *Harvard College Observatory*

Focused by magnetic fields, protons and electrons in nearly equal numbers reach the earth and, as Gartlein and Meinel have shown, produce broadened and shifted lines of the Balmer series in the aurora borealis. An astrophysical analysis of the emission of hydrogen gas in the neighborhood of the earth indicates that the number of protons and electrons per cm^3 of the gas responsible for this emission is of the order of 10^6 .

An analysis of the effect that gas clouds would have on the earth's magnetic field indicates that the latter can exert a focusing action. The corpuscular solar radiation responsible for the aurora borealis tunnels its way through the magnetic barrier of the earth's field.

The total energy carried by the ion stream may approach that of solar radiation itself. Thus, we can relate corpuscular radiation to the heating and dynamics of various portions of the earth's atmosphere.

Technetium in the Stars

Paul W. Merrill

Mount Wilson and Palomar Observatories

Technetium, the first "artificial" element, was identified in 1937 by Perrier and Segrè in a piece of molybdenum that had been bombarded with neutrons in the cyclotron at Berkeley. Technetium has also been detected among the products of fission of heavy atoms. No completely stable isotope is known; the most nearly stable has a half-life less than a million years.

The spectrum of technetium was thoroughly investigated in 1950 by Meggers and Scribner at the National Bureau of Standards. Their work has made astronomical investigations possible. In 1951 Charlotte E. Moore announced the possible presence of weak lines of ionized technetium in the solar spectrum.

Spectrograms, dispersion 10 Å/mm, taken by P. W. Merrill with the 100-inch telescope and others taken recently by I. S. Bowen with the 200-inch, show several lines of neutral technetium in the spectra of S-type stars, especially of long-period variables. The strongest of these lines are in the $a^3S - x^3P$ multiplet, analogous to the well-known triplet at $\lambda 4030$ in the spectrum of manganese. Stellar spectra of type S are characterized by bands of zirconium oxide and by relatively strong lines of heavy metals such as zirconium and barium.

It is surprising to find an unstable element in the stars. Either (1) a stable isotope actually exists although not yet found on earth; or (2) S-type stars somehow produce technetium as they go along; or (3) S-type stars represent a comparatively transient phase of stellar existence.

Methods of Using Binoculars

Walter R. Miles, *Yale University*

There are two basic methods of using binoculars: (1) keeping them continuously at the eyes and (2) locating the field by naked eye and then using the binoculars. The relative advantages of the two methods were examined on a group of U. S. Navy personnel and a control group matched for age and visual qualifications. Five target areas, $\frac{1}{2}$ –2 miles distant, seen against the sky, were used. Observation was from a rooftop station. Standard 7×50 binoculars were employed. The targets, varying in size and contrast, were each made of two black or gray bars that could be oriented vertically or horizontally and were presented in a randomized series determined for each subject individually. Subjects were tested individually after pretraining, and each man made 115 response judgments. The continuous method (1) was found to give more prompt judgments in both groups by 0.2–0.3 sec. In terms of threshold resolution the discontinuous method (2) proved superior by about one log step for target size and target contrast. The choice of method, therefore, in particular military situations should be in terms of the anticipated hazards.

In designing the targets it was assumed that the 7×50 binoculars with coated lenses might yield 4.5 efficiency over naked eye performance. The results indicate that the assumption was more than reached and that the probable efficiency of such glasses over the naked eye is 5:5.5 in such critical tests and distances as were used in these daytime experiments.

Recent Determinations of Atomic Masses and Nuclear Binding Energies

Alfred O. C. Nier, *University of Minnesota*

A double-focusing mass spectrometer has been developed which, because of its sensitivity and versatility, makes possible the measurement of atomic masses of virtually every isotope of any element. A systematic study of two regions of the atomic table has been completed, from S^{32} to Zn^{60} , where 42 of the 45 naturally occurring nuclides have been measured, and from Pd^{100} to Xe^{136} , where measurements on 42 of the 48 stable nuclides were made. Measurements are made by the familiar doublet method, hydrocarbon fragments serving as comparisons.

The mass-spectroscopically determined masses, when supplemented by nuclear reaction data, furnish a large block of data making possible extensive comparisons with semiempirical formulas for nuclear binding energies. A comparison with the formula of Wigner indicates discontinuities in the binding energy surface at 20 and 28 protons and neutrons and at 50 protons. The data are sufficiently accurate to reveal other less pronounced details of fine structure.

Low Temperature Properties of Helium 3

Darrell W. Osborne, Bernard M. Abraham, and Bernard Weinstock, *Argonne National Laboratory*

Sufficient quantities of pure He^3 have been obtained by nuclear transformations to permit investigations of

some properties of the liquid. These investigations are of interest because they may aid in the interpretation of the extraordinary properties of liquid He^4 . By studying the flow of the liquids through a small channel it was found that He^3 , in contrast to He^4 , does not exhibit the phenomenon of superfluidity. This result is consistent with the theory that superfluidity is connected with the fact that He^4 follows Bose-Einstein statistics, whereas He^3 follows Fermi-Dirac statistics. It has also been shown that the addition of He^3 to He^4 depresses the temperature below which the liquid behaves as a superfluid, and that the transition to the superfluid state remains of the second order type as He^3 is added. Further, the viscosity of liquid He^3 , like that of all other liquids except He^4 , was found to increase as the temperature is lowered. Measurements of the melting pressure of He^3 between 1.5 and 0.16° K have shown that He^3 , like He^4 , has the remarkable property of remaining liquid when cooled to absolute zero, unless a high pressure is applied. Above 0.5° K the data are represented by the equation $P = 26.8 + 13.1 T^2$ atmospheres, but below 0.5° K the experimentally determined melting pressures rapidly approach a constant value of 29.3 atmospheres. The results below 0.5° K may be spurious because of possible poor thermal contact between the capillary containing the He^3 and the paramagnetic salt used for cooling below 1° K, but if they are correct they imply a transition in the liquid near 0.5° K. Further experiments are in progress to clarify this point.

Common Metabolites and Stimulation or Inhibition of Growth by Plant Tissue Cultures

A. J. Riker and A. C. Hildebrandt
University of Wisconsin

Various plant tissues have been studied *in vitro* to clarify factors that stimulate or inhibit growth. They came from crown gall on marigold, *Paris* daisy, periwinkle, and sunflower, and from normal hybrid tobacco stems. They grew during 10 years through many transfers on synthetic media. The amount of growth after six weeks was determined by weighing the tissue pieces. The results reported came from over 60,000 pieces of tissue.

Common metabolites, depending on their concentrations, respectively, often encouraged or discouraged growth of the various tissues.

With various sources of nitrogen at pH 6.0, growth was excellent with nitrate (.008 M) and urea (.032 M). Except on arginine, growth of sunflower tissue was sharply inhibited by all the amino acids tried at .001 M. However, satisfactory growth developed with relatively strong (.004 M) concentrations of alanine, aspartic acid, and glutamic acid, possibly because of transamination.

One tissue or another, respectively, used a wide range of sugars and polysaccharides. Some showed best growth with certain substances at particular concentrations.

The common alcohols at pH 6.0 were poor as sole sources of carbon. With 2% sucrose, some common alcohols inhibited specific tissues at particular concentrations. For example, all five kinds of tissue were inhibited by butanol at .125%, and by propanol at .06%.

The common organic acids at pH 6.0 were poor sources of carbon. With sucrose, many of them inhibited growth. For example, marigold tissue was inhibited by 0.03% of propionic, glutaric, and acetic acids.

Nondilation of Arteries with Pulsating Blood Flow

S. R. M. Reynolds, *Carnegie Institution of Washington*

In the course of collaborative studies of the fetal circulation with G. M. Ardran and M. M. L. Prichard, of the Nuffield Institute for Medical Research, Oxford, observations were made which show that there is no lateral or radial deformation of arteries associated with physiologically pulsating blood flow. In umbilical arteries, direct cineangiographic pictures of blood flow were recorded at 25 frames/sec. Thorotrust was injected into the aorta by way of an iliac artery. Blood pressure was recorded from a branch of the umbilical artery. The following facts were established: (1) blood flow in the artery is laminar; (2) flow is pulsatile; (3) there is no detectable lateral deformation of the artery observed (a) in projected moving pictures or (b) in tracings of arteries in successive frames in a series; (4) there is lateral pulsatile displacement of the entire artery in certain regions, especially in the vicinity of curves where there is deflection of the pulsating streamline flow.

The fact that pulsating arteries do not dilate but are displaced locally was confirmed throughout the descending aorta of the cat by direct cineangiography. Only the ascending aorta and the arch of the aorta expanded and contracted with periodic ejection of blood from the heart.

Contrary to common belief, pulsatile arterial blood flow is not associated with significant radial deformation of the vessel wall in the propagation of a pulse wave. It therefore appears that the pulse wave is transmitted as a wave of pressure within the moving blood stream.

Nature of Inherited Resistance to Viruses Affecting the Nervous System

Albert B. Sabin
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The accidental discovery that an inbred line of mice (PRI) is 100% resistant to the 17 D strain of yellow fever virus made it possible for the first time to work out the mechanism by which animals inherit resistance to certain viruses. The results indicate that resistance to the 17 D virus is inherited as a dominant in accord with the Mendelian laws for a single pair of autosomal genes. The dominant gene does something to the cells of the resistant host to make them a poorer medium for the propagation of yellow fever virus—multiplication being 10,000–100,000 greater in the brains of susceptible than in those of resistant mice. The lower level of viral multiplication permits the development of immunity without disease.

The possession of the dominant gene does not, however, protect the mice during the first few days of life. The virus does not multiply to a higher level in the newborn, but its cells are more vulnerable. The mouse-adapted French neurotropic strain of yellow fever virus kills 24% of adult PRI mice, even though it multiplies only to the same low level as the 17 D virus. The special cellular vulnerability to the French neurotropic strain of yellow fever was also found to be genetically determined, but independently of the factor controlling viral multiplication.

The genetic factor which depresses multiplication of

the yellow fever virus has a similar effect on the viruses of dengue, West Nile fever, Japanese B, St. Louis, and Russian tick-borne encephalitis, but is without effect on the viruses of western equine, eastern equine, and Venezuelan encephalitis, poliomyelitis, rabies, and a number of others tested. It was also found that some viruses possess or can develop variants which can overcome the genetic resistance of the host.

Cloud Chamber Observations of the New Unstable Particles in Cosmic Rays

Bruno Rossi, *Massachusetts Institute of Technology*

The MIT group has operated for several years a multiple-plate cloud chamber at the Inter-University High Altitude Laboratory at Echo Lake, Colo. (alt., 10,600'). Most of the time the chamber was triggered by a penetrating shower selector placed above it. This triggering arrangement afforded the possibility of an unbiased investigation into the properties of the secondary particles from nuclear interactions occurring above the chamber. Among these secondary particles we found many examples of the new unstable particles. Some of these decay in flight and exhibit the characteristic appearance of the so-called V-particles (both charged and neutral); the large amount of material present in the chamber made it possible to investigate, in some detail, the nuclear interactions of the secondary particles arising from their decay. Other particles disintegrate after coming to rest in matter. Their secondary products are heavier than electrons, for they traverse several lead plates without multiplication. These particles, for which our observations provided the first experimental evidences, are possibly identical to the so-called κ -particles that shortly afterward were detected in photographic emulsions by the Bristol group. Our results, together with those of the Bristol group, make it appear unlikely that κ -particles may be identical to charged V-particles.

Imperfections in Crystals: A Synthesis

Frederick Seitz, *University of Illinois*

The primary imperfections responsible for many of the most interesting properties of crystals have been studied in detail for about 25 years. During this period separate imperfections have been introduced into the field to explain individual properties, such as plasticity, diffusion, electrical resistivity, and many other characteristic properties. At the present time six primary types of crystal imperfection are recognized: phonons, electrons and holes, excitons, vacant lattice sites and interstitial atoms, foreign atoms in either interstitial or substitutional position, and dislocations.

It is regarded as unlikely that there are more primary imperfections characteristic of the principal types of solid, although special imperfections may exist in materials with specialized properties, such as ferromagnetic materials. In addition to the primary imperfections there are three transient imperfections: light quanta, charged radiations, and uncharged radiations. The transient imperfections may have a brief life within the crystal. The individual imperfections are discussed with a brief account of the historical introduction of each type. It is pointed out that two or more imperfections of the same type or of different types may interact to generate other imperfections, so that the group forms a closely interlocking family. This feature of the imperfections is regarded as an essential part of their pattern of behavior,

intimately related to the normal properties of the imperfections.

Seismic Results Relative to Crustal Structure in the Wisconsin Area

Louis B. Slichter, *University of California*

Seismic results from six large underground blasts in the Wisconsin area are reported. The observations were made with a group of a dozen three-component seismographs, located on selected bedrock sites. Time signals at 1-sec intervals were transmitted by radio. On the scale of the present work, which involved an area about 600 km \times 150 km, the major structure was a relatively homogeneous layer about 40 km thick. The observed time-distance relations corresponding to this layer are nearly linear and would imply constant compressional wave velocities of 6.16, 6.26, 6.22, and 6.16 km/sec, respectively, at the four regions investigated. However, a somewhat improved fit to the observational data is obtained if the wave velocity is not constant, but increases slightly from 6.0 km/sec at the top to 7.0 km/sec at the bottom of the layer. Under the latter assumption, the mean absolute value of the time residuals is reduced to about one half its value for the assumption of a homogeneous layer. Above the main layer is a thin superficial layer 0.6-2.8 km thick, with wave velocity 4.2-4.6 km/sec. The coarse scale of the field observations permits only spotty sampling of this thin upper layer. Beneath the thick, nearly homogeneous layer, at depth 40-44 km below the ground's surface, a major crustal discontinuity is indicated, where the wave velocity increases suddenly from 7.0 km/sec above the transition to 8.2 km/sec below.

The Action of Isotopic Insulin Bound to Tissues

William C. Stadie, Niels Haugaard, and Martha Vaughan *University of Pennsylvania*

We previously reported that muscle, liver, adipose tissue, and lactating mammary glands of rats bind insulin. The bound insulin exerts its customary effect on metabolism.

In further work we prepared isotopic insulin labeled with S^{35} and I^{131} to relate biological effect to mass of insulin. These preparations have greatly extended the precision and significance of our experiments. Some types of experiments which will be reported are: (1) Insulin bound is a function of concentration and time of exposure. (2) There is a saturation level of bound insulin at higher concentrations of insulin. (3) Insulin binds readily at pH 2. (4) The binding is increased with rising temperature. (5) There is a close correlation between insulin bound and its biological activity. This was measured both *in vivo* and *in vitro* with concordant results. These measurements for the first time measure the biological activity per unit mass of a hormone. (6) Measurements of distribution of insulin following intravenous and subcutaneous injection show contrasting results.

These experiments lead to the concept that a prerequisite for insulin action is a chemical binding on intact cellular elements. Contra-insulin effects of pituitary or adrenal hormones may either: (a) inhibit the action of the bound insulin, or (b) inhibit the binding of insulin. In either case the metabolic activity of insulin would be decreased but by quite different mechanisms.

The distinction between these two possibilities is of fundamental importance in the problem of hormonal interaction.

Further Studies on the Localization of X-Ray Injury to the Initial Phases of Antibody Response

W. H. Taliaferro, Lucy Graves Taliaferro, and E. F. Janssen, *University of Chicago*

Rabbits have been injected intravenously once with 1.25 ml of 1% sheep red cells at intervals from 4 days before to 38 days after a single total body x-irradiation with 600 or 700 r. Hemolysin was determined photometrically in 50% units at frequent intervals. Maximum inhibition of antibody production occurred in rabbits receiving antigen 1-7 days after irradiation, and consisted of doubling the induction period and reducing peak titer to one seventh that in nonirradiated controls. Ability to form normal amounts of antibody was recovered in most animals receiving antigen 28 days after irradiation. The most striking finding was that animals receiving antigen from 2 days before to 3 min before starting irradiation formed normal amounts of antibody, although many exhibited lengthened induction period. X-ray injury is probably largely limited to an early phase of antibody formation, because in the last group (1) the most pronounced injury was a prolongation of the induction period, and (2) normal amounts of antibody were formed in the week following x-irradiation when x-ray injury is most apparent. This critical phase may be a stage in antigen localization or metabolism. It is related to the amount of antigen injected since, in another series of experiments, increased antigenic stimulation given as a series of injections also stimulated the formation of normal amounts of antibody after a lengthened induction period, even though given during the week following irradiation.

Systematic Errors in Elemental Abundances in Meteorites

Harold C. Urey, *University of Chicago*

It has previously been suggested that the chondritic meteorites are an average sample of nonvolatile planetary material. These objects, judging from their structure, have been heated to about the melting point of iron for brief periods only. The question arises as to whether certain elements have been markedly volatilized from them, thus leading to systematic errors in elemental abundances. The volatility of compounds of the halogens, sulfur, phosphorus, the alkalis and alkaline earth metals, and silicon leads to the conclusion that none have been markedly fractionated, but that halogens would be lost rapidly from cosmic mixtures heated to these temperatures, and that iron oxides and sulfide would be rapidly reduced in hydrogen gas. Since bromide and iodine have been retained, no great losses of any of the halogens have occurred. It is concluded that these objects were made rapidly in times of the order of one day or less. A collision between two asteroids with relative velocities of some two km/sec, such as has often been suggested, would supply adequate temperatures and would last perhaps some 10 min. Objects resulting from such a collision would cool rapidly to low temperatures.

From a reconsideration of volatilities of sodium, magnesium, calcium, and silicon oxides in the presence of cosmic gases, it is concluded that the first mechanism proposed by the writer for the fractionation of the iron and silicate phases as the earth and other terrestrial planets were formed—i.e., by differences in volatility—cannot by itself explain this effect and that the second

method proposed—namely, by iron sinking below a molten silicate layer and thus not being volatilized while the silicate layer was rapidly evaporated—is a required process.

Studies of Populations Exposed to Radiation

Bruce Wallace

Biological Laboratory, Cold Spring Harbor

Experimental populations of *Drosophila melanogaster* exposed to continuous γ -radiation or to a single, massive dose of x-radiation are being studied. The analyses are primarily of two types: determination of the frequencies of second chromosomes carrying lethal and semilethal gene mutations in the several populations, and estimation by various techniques of the adaptive values or well-being of the populations. Two populations (the control and the one exposed to x-rays) have been maintained for over 120 weeks (approximately 60 generations), and three others (those exposed to γ -rays) for about 80 weeks.

Gene content, as revealed by the second-chromosome analyses, varies greatly among the five populations. Lethal-chromosome frequencies range from about 15% to about 70%; semilethal-chromosome frequencies from about 5% to about 20%. The average viability of flies homozygous for nonlethal, nonsemilethal second chromosomes is relatively constant within any one population (even those receiving continuous radiation), but each population has its characteristic frequency.

The estimates of adaptive value vary, depending on the technique used for estimation, but nevertheless present a consistent pattern, which indicates a lower value in those populations that have received the most chronic irradiation.

The results indicate that the genetic structures of populations are under constant review by natural selection, and that well-adapted structures are maintained in spite of mutagenic forces operating counter to natural selection.

Self-Regulation of Organ Growth by its Own Products

Paul Weiss, *University of Chicago*

In 1945, the author proposed a concept of organ-specific growth control as follows. Protoplasm synthesis of a given organ yields (a) "templates" for further reproduction, and (b) accessory diffusible compounds capable of inactivating the former. As the latter, accumulating in the common humoral pool, reach critical concentration, growth ceases. Partial removal of an organ, by reducing the concentration of (b) will automatically entail "compensatory" growth in the rest of the organ. Growth and differentiation, being somewhat antagonistic, may react in opposite directions.

The organ-specific aspects of this concept have now been subjected to three new experimental tests. In one (with I. Fischer), embryonic chick organs were reared in tissue culture media prepared with extracts of whole embryos and of embryos from which the homologous organ had been removed. Of 333 heart fragments with healthy growth in full extract (2-day transfers), only two pulsated after the fourth day, whereas of 349 fragments similarly reared except for omission of heart extract from the medium, 129 pulsated, indicating progressive muscular differentiation. Likewise, of 1007 embryonic kidney fragments (metanephros) in full extract,

only 74 differentiated new tubules in the growth zone, as against 176 of 1006 cultures grown without kidney extract. Presence of a given organ extract thus markedly reduces differentiation capacity in the homologous organ.

In another test (with G. Andres), embryonic kidney brei was injected intravenously into younger embryos. Mitotic counts after 24 hr showed a 50% increase above normal in the host kidneys, with electivity of the effect indicated, though not yet crucially proved. In a third test (S. Ferrin), the destruction of one embryonic metanephros in a prefunctional stage entailed "compensatory hyperplasia" of the remaining kidney (12 cases) marked by a more than 100% increase of mitotic activity within 48 hr. Functional overload being excluded, this result corroborates the operation of an automatic growth control principle such as outlined above.

Winds in the Upper Atmosphere by Meteor-Train Photography

Fred L. Whipple, *Harvard College Observatory*

A new technique for the study of persistent meteor trains and the measurement of wind velocity in the upper atmosphere has been made possible by the development of the Baker Super-Schmidt meteor camera. This instrument, operating at $f/0.65$ optically, with an aperture of 12 in. and a field of 54° , can obtain multiple photographs of the persistent luminous trains left behind very bright meteors. The objective shutter is opened on the appearance of the meteor, and a programming device is used to shift the telescope slightly at intervals of 2 sec. The first Super-Schmidt meteor camera, belonging to the U. S. Naval Bureau of Ordnance, located at one of two meteor-observing stations near Las Cruces, N. M., has been operated by the Harvard Observatory for some months under a contract with the U. S. Office of Naval Research. After considerable experimentation with photographic emulsions and processing, Robert C. Wells obtained the first successful multiple photograph of persistent train with the new equipment on the night of December 22, 1951. The meteor itself was photographed by small cameras at both stations. A persistent train extended from an altitude of 94 km to 81 km above sea level. The northwest-southeast component of the wind vector normal to the meteor trail remained usually less than ± 50 km/hr except for a 150-km/hr current of 2-km depth centered at an altitude of 88 km. The wind component changed by about 150 km/hr from southeast to northeast in an altitude range of only 1700 feet. The precision of wind measurement is about 5 mph over an altitude range of 0.1 km. The highly turbulent character of upper atmospheric circulation is indicated clearly in these results. The decay rate of the train increased markedly with decreasing altitude.

Routine systematic photography of persistent meteor trains by the new technique is planned to begin in the fall of 1952, when two Super-Schmidt meteor cameras will be put into operation in New Mexico for the U. S. Air Force.

The Giant Mitochondria of Insects

Carroll M. Williams and Mary I. Watanabe
Harvard University and U. S. Army Quartermaster Research and Development Laboratories

The striated muscles that propel the wings of the higher insects (Diptera and Hymenoptera) are clearly

divisible into two mechanisms: a contractile mechanism localized in the actomyosin of the fibrils, and an energy-trapping mechanism centering in the sarcosomes. The latter are spherical, intracellular bodies, $1 \mu - 6 \mu$ in diameter, aligned in individual longitudinal rows between the fibrils. They make up one third of the muscle mass and are readily isolated by centrifugation or filtration.

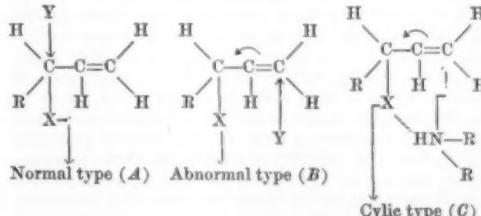
Cytological, histochemical, spectroscopic, and biochemical studies demonstrate that the sarcosomes are the "giant mitochondria" of this extraordinary muscular tissue. Within the muscle fibers the sarcosomes appear to be the principal sites of oxygen consumption, electron transmission, and ATP formation. The succinoxidase system is localized exclusively in these bodies.

Each sarcosome consists of a gelatinous matrix surrounded by a discrete membrane. The latter is disrupted by freezing and thawing, by temperatures above $40^\circ C$, by the vapors of fat solvents, or by the addition of digitonin; the contents are released as minute granules. The sarcosomal membrane is permeable to water, electrolytes, ATP, intermediates of the Krebs cycle, pentoses, and phosphorylated hexoses. But it is essentially impermeable to protein, AMP, and nonphosphorylated hexoses. Substances that penetrate the sarcosome exert only transient osmotic effects on the membrane. The osmotic entry of water then results in a pronounced swelling of the sarcosome. Its contents are frequently extruded, leaving the empty membrane as a discrete, spherical "ghost."

The Abnormal Bimolecular Displacement Reaction in Allylic Systems

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Two types of second-order nucleophilic displacement upon allylic halides are recognized:



The most common type (A) is a normal nucleophilic displacement (S_N2) upon the saturated α carbon atom bearing the leaving group, producing an allylic system identical with the original, except group Y has replaced group X . The second type (B) is an abnormal nucleophilic (S_N2) displacement upon the unsaturated carbon atom γ in the original system, accompanied by rearrangement of the double bond and expulsion of the halide, and producing a new allylic system in which the entering nucleophile Y has become bonded to the carbon atom γ in the original system. All unambiguous cases of S_N2 displacement upon allylic halides involving amines as the entering nucleophile have been demonstrated with dialkyl amines. In these cases the possibility exists that the success of S_N2 displacement over S_N2 displacement is due to the attraction between the amine hydrogen atom and the halide atom (hydrogen bonding), followed by a cyclic reaction type (C). It has now been shown that hydrogren

bonding is not essential to the success of the S_N2 displacement by amines on allylic halides by employing the reaction of trimethyl amine (which has no hydrogen atom to engage in hydrogen bonding) with α -methylallyl chloride in acetone. Both normal and abnormal products are formed by S_N2 and S_N2' displacements, respectively. No rearrangement of starting material or products occurs. The kinetics are second order. Heats and entropies of activation have been calculated for both the S_N2 and the S_N2' displacements.

Intellectual Ability of Students Entering Different Fields of Science

Dael Wolfe

Commission on Human Resources and Advanced Training
National Research Council

High school grades. Of 1050 students who earned Ph.D.'s in science, 21% were high school valedictorians and 62% were in the top 10% of their classes. The fields ranged from physics and mathematics, with 77%, to agriculture, with 48%, from the top 10%.

Scholastic aptitude tests. Among college graduates, average scores for engineering, language, physical science, and psychology majors are above college averages. Biological science, social science, and arts majors about equal college averages. Agriculture, business, and education students are below average.

Twelve per cent of science Ph.D.'s were in the top 1% and 52% in the top 10% of college freshmen. Percentages from the top 10% are: physical sciences, 60; earth sciences, 52; biological sciences, 40; and agriculture, 26.

These differences disappear among the most eminent research scientists. Roe found that eminent physicists, biologists, and psychologists and anthropologists all averaged far above the typical Ph.D., but tapered off to an ability level only moderately above the undergraduate average. Interest and persistence can produce

research eminence from a wide range of intellectual ability, but typically the most eminent come from among those who make very high test scores. The distribution of such men among the sciences is uneven.

The Anomalous Transparency of Thick Crystals to X-Rays

W. H. Zachariasen

Argonne National Laboratory and University of Chicago

The anomalous transparency of some thick crystal specimens to a beam of x-rays producing "Laue case" diffraction was first observed by Borrman. Quantitative experimental studies of the effect, using calcite crystals, have recently been made by Borrman and by H. N. Campbell.

The complete solution for Laue case diffraction in absorbing perfect crystals is capable of explaining the observed phenomenon. In order to make a quantitative comparison between theory and experiment it has been necessary to modify the solution so as to fit the specific experimental procedures used by Borrman and by Campbell. This modification has been made, and the result is presented in the form of an expression for the effective absorption coefficient.

The usual absorption coefficient of calcite for x-rays of wavelength 1.54\AA is 193 cm^{-1} . The experimentally measured and the theoretically calculated values for the effective absorption coefficient in the region of anomalous transparency are given in the table below.

Crystal thickness (cm)	Calculated (cm^{-1})	Measured (cm^{-1})	Experimenter
0.040	79	86	Campbell
.212	30.5	30.2	Borrman
0.271	28.6	27.7	"

William de Berniere MacNider: 1881-1951

W. C. George

Department of Anatomy, School of Medicine, University of North Carolina, Chapel Hill

WILLIAM DE BERNIERE MACNIDER, son of Virginius St. Clair and Sophia Beatty (Mallett) MacNider, was born in Chapel Hill, North Carolina, June 25, 1881, and died in Chapel Hill, May 31, 1951. He received his elementary education in the schools of his home town, and his college and professional training at the University of North Carolina. After three years of study in college and the preclinical Medical School at Chapel Hill he transferred to the clinical school of the university that had just been established at Raleigh. He graduated there with the first class of young men to receive the degree of Doctor of Medicine from that institution. His professional education was

extended during subsequent summers at the University of Chicago and Western Reserve University.

Throughout most of his life Dr. MacNider was a member of the faculty of the University of North Carolina, being successively assistant in biology (1899-1900), assistant in anatomy (1900-1902), assistant in clinical diagnosis (1902-1905), professor, Kenan professor, and Kenan research professor of pharmacology (1905-1950). He retired as professor emeritus in 1950. From 1937 to 1940 he was dean of the Medical School.

Dr. MacNider's achievements and the force and charm of his personality were recognized by honorary degrees from Davidson College and the Medical Col-

lege of Virginia, by memberships and official posts in scientific and social groups, and by medals and lectureships. He was a member of the American College of Physicians, the National Academy of Sciences, the American Philosophical Society, and many other scientific organizations. He served as president of the American Society for Pharmacology and Therapeutics, the Medical Society of North Carolina, the Elisha Mitchell Scientific Society, the Society of Experimental Biology and Medicine, the International Anaesthesia Research Society, and the Gerontological Society. He was awarded the Gibbs prize of the New York Academy of Medicine, the Research Medal of the Southern Medical Association, and the Kober Medal of the Association of American Physicians.

Dr. MacNider's principal contributions to creative scholarship may be grouped into five categories: (1) The production of various forms of acute and chronic nephritis, with cytological study of the types of injury and of the processes of repair; (2) disturbances in the acid-base balance in the blood in experimental nephritis during anesthesia and intoxication by salts of heavy metals; (3) the influence of lipid accumulations within renal cells upon the susceptibility of the cells to the toxicity of anesthetics, together with demonstration of the ability of alkalies or glucose to decrease that susceptibility; (4) studies of the factors of age in the response of animals to normal and abnormal influences; and (5) evidence that, when certain epithelial tissues are severely injured by toxic agents, the succeeding repair process forms a typical, yet functionally effective, kind of cell with an acquired resistance to the chemical agent of injury and other agents of different chemical order.

As the son and grandson of a physician, and with a home in Chapel Hill at a time when the ferment of modern science was becoming active there, Will MacNider could attribute his interest in medical science to heredity and environment. As a boy he was interested in nature, and as a young undergraduate he acquired an enthusiasm for science. He once said that his enthusiasm was sparked by the study under the microscope of a living kidney tubule of an earthworm. Many another young student has been thrilled by the sight of life in operation on the cellular level, but for young MacNider the thrill was seed in fertile ground. The spectacular sight of the earthworm's nephridium fired him with a passion to penetrate the secrets of organs, of cells, of tissue fluids, of organisms—to devote a lifetime of research principally to a study of the function and pathology of the kidney. Although this line of research was never abandoned, his interests broadened to encompass an interest in the fundamental nature of the vital processes of organisms and their philosophical implications.

He was fortunate in early coming under the stimulating influence of some of the inspiring pioneers of science of fifty years ago. H. V. Wilson and Richard Whitehead, of Chapel Hill, S. A. Matthews, of Chicago, G. N. Stewart and Torald Sollmann, of Cleveland, and Thayer and Osler, of Johns Hopkins University, were men with whom he was associated, and from whom he drew inspiration and stimulus during his early years. His own eagerness and industry in the exciting search for new truth contributed largely to the intellectual climate of his own university during the fifty years that he was a part of it.

The list of his publications supplies ample testimony to his untiring activity and his wide interests. His contributions to science and teaching are on deposit in the medical library. They consist of twenty volumes of bound reprints, a gift from him to the library about a year before his death. His interest in the library was further demonstrated from time to time by gifts of books and journals, by the gift of his extensive collection of reprints from investigators all over the world, and by the bequest in his will of the scientific books remaining in his considerable library.

Although he valued highly the friendship and acclaim of his professional colleagues and contemporaries, his greatest reward was the esteem and affection in which he was held by students that passed under his tutelage. His zest for learning could not make a recluse of him, because it was accompanied by an equal zest for companionship that drew students to him in lifelong friendships.

Research and teaching occupied most of his time and energies, and yet enough was left over for a live and active interest in his home, which was the center for much pleasant hospitality. Work in his garden filled the place that sports fill in many men's lives. Gifts of flowers to neighbors and local friends were many and frequent, and many acquaintances throughout the country have had their hearts gladdened and their gardens beautified through gifts of flower seeds from his garden.

Throughout his career he remained profoundly interested in the methods and problems of medical education and in the practical, as well as the philosophical, implications of scholarship. In the council chamber and committee room he was a forceful exponent of his views, and his opinions were sought and weighed with interest by antagonists, as well as by supporters.

Although intensely interested in his own thoughts and his own affairs, Will MacNider had a keen interest in the work, the pleasures, and the sorrows of those with whom he came in contact. He contributed to their pleasures and knew how to be of help in their sorrows. His death has left a gap in the domain of science and in the lives of hosts of friends.

News and Notes

Scintillation Counter Symposium

A SYMPOSIUM on the rapidly developing field of scintillation counters was held under the joint sponsorship of the National Bureau of Standards, the Atomic Energy Commission, the American Institute of Electrical Engineers, and the Institute of Radio Engineers January 29-30, at the Shoreham Hotel, Washington, D. C. The meeting was attended by more than 300 scientists representing universities and government and industrial laboratories.

The symposium was divided into four sessions, under the respective chairmanships of L. S. Taylor (NBS), G. A. Morton (RCA), W. H. Jordan (ORNL), and J. B. H. Kuper (BNL). During the two-day period, 35 papers were delivered, some of which are briefly summarized here. In his welcoming address, A. V. Astin, acting director of the National Bureau of Standards, expressed the interest of the bureau in the development of new techniques of measurement such as the scintillation counter.

The opening paper, delivered by M. H. Greenblatt (RCA), was devoted to a description of two new multiplier phototubes developed in the RCA laboratories by P. W. Davison, G. A. Morton, M. W. Green, and the speaker. The first of these tubes (RCA H-5037) has a 3.5" photoanode for use with large scintillation materials. It contains an electrostatic focusing system and a conventional 931-A multiplier structure. The second of the tubes (RCA 4646) has extremely high gain ($\sim 10^9$), eliminating in most cases the need for after-amplification. This is accomplished through use of a 16-stage dynode-anode structure. The very high gain of the tube introduces a number of space charge problems; however, output pulses of 40-v amplitude into a 200-ohm line are obtained easily with conventional phosphors. Similar tubes developed at EMI were described by J. McGee (England). The British tubes are of the Venetian blind type of structure.

Several relatively new instruments and circuits were discussed. C. H. Smith (NRL) described a cathode-ray tube capable of registering 10^{11} sweeps/sec, having a deflection sensitivity of 0.6 v/trace width. This instrument utilizes a traveling wave deflection system. Two very high resolution coincidence circuits were described. One developed by J. Marshall (Chicago) employs a type 6BN6 gated beam tube and has a resolution time of the order of 5×10^{-10} sec. The other, described by Z. Bay (Washington U), makes use of germanium diodes in a differential circuit and has a resolving time of the order of 2×10^{-10} sec.

High-speed circuits using secondary emission pentodes (EFP60) and very high transconductance pentodes (VX 5038) were reported from England by F. H. Wells (AERE). In further discussions of circuits, G. G. Kelley (ORNL) indicated the difficulties en-

countered in the design of pulse amplitude analyzers for scintillation counter spectrometry, and W. A. Higinbotham (BNL) analyzed new electronics required for optimum use of scintillation counters.

Serious limitations in the application of scintillation counters to high resolution spectrometry and very high resolution coincidence counting are imposed by satellite, or after-, pulses and by the finite emission time and transit time in the multiplier. Various aspects of these problems were presented by D. W. Mueller (LASL), F. B. Harrison (Princeton), D. C. Moore (RCA), P. W. Davison (RCA), R. W. Engstrom (RCA), R. R. Law (RCA), G. A. Morton (RCA), and R. F. Post (California). Small pulses occurring within a few microseconds following a single event have been observed. The amplitude of these satellite, or after-, pulses is about twice that of the multiplier noise pulses. They are probably caused by the formation of positive ions in the multiplier which produce secondary electrons in the photocathode. Effective elimination is possible by careful outgassing of the multiplier; this, however, requires the use of special dynode materials.

It was pointed out by R. R. Law that the time spread in the electrons arriving at the anode is attributable in part to finite electron emission time. This was determined experimentally through use of a secondary electron multiplier structure of extremely small dimensions (1 mm).

Work on phosphors is oriented toward the development of higher efficiency, higher speed, sensitivity to slow neutrons, and larger effective volume. It appears that for γ -ray detection NaI(Tl) is the most efficient phosphor in the low-energy region, and the organics such as anthracene and stilbene at higher energies. The shortest decay times are found in liquid scintillators such as terphenyl in toluene.

The inclusion of boron or lithium in the phosphor permits the detection of slow neutrons by the (n, α) reaction. J. C. Schenk (ORNL), R. K. Swank (Argonne), and A. W. Schardt (BNL) discussed the performance of various boron- and lithium-containing phosphors. The most promising of the materials is LiI (Tl), which shows a narrow pulse height distribution originating from the α -particles produced in the (n, α) reaction.

R. C. Sangster (MIT) described experiments on the relationship between the fluorescence spectrum and the structure of systems of condensed ring compounds.

Investigations of the fundamental processes of energy transfer in organic fluors were reported by F. N. Hays (LASL) and S. H. Liebson (NRL). Dr. Hays' experiments involved the use of compounds containing C^{14} and H^3 . The evidence found is consistent with a theory that involves initial ionization steps, followed by energy transfer from solvent cations to solute molecules producing solute cations,

which recombine with low-energy electrons to give activated scintillator molecules, subsequently returning to their ground state with simultaneous emission of light.

Dr. Liebson's work was concerned with the behavior of anthracene-naphthalene mixtures as a function of temperature. The results indicate that the anthracene in naphthalene acts as an energy trap, the host lattice being primarily responsible for the temperature variation of the pulse size. Several different methods for the measurement of fluorescence decay time were also discussed. Differences in decay time for ultraviolet excitation as compared to γ -excitation are indicated by these measurements.

In the region of the very high energies the application of conventional scintillation counters was described by L. F. Wouters (California). Another technique that appears promising for the measurement of electrons, mesons, and protons in this region is the detection with a multiplier tube of Cerenkov radiation from various media. J. Marshall (Chicago) discussed the problems involved in the application of this method.

Cosmic ray investigations employing very large volumes of liquid scintillators viewed by several multiplier tubes were described by G. T. Reynolds (Princeton) and F. H. Wells (Harwell). A similar application of liquids to the measurement of the energy distribution of the beam from the NBS betatron was presented by Cleland and Koch (NBS).

Pulsed operation of a photomultiplier can sometimes be employed to maximize its gain, output current, and speed of response. In a paper given by R. G. Post (Stanford) a practical case using a standard 1P21 photomultiplier was described. It was found that under pulsed conditions the multiplier gains were sufficiently high ($\sim 10^9$) to enable the direct display of noise output pulses having a width of 10^{-9} sec or less, on a cathode-ray tube. Experiments were carried out in the use of this technique for very fast coincidence counting, with resultant resolving times of better than 10^{-9} sec.

With a simple coincidence circuit having a resolution of about 10^{-9} sec, S. De Benedetti (Carnegie Tech) was able to measure the time of flight of γ -rays over distances of the order of a few centimeters. He applied this instrument in a study of the half-life of positrons in condensed materials and in the measurement of the α - γ angular correlation of Po^{210} .

One of the most fruitful applications of scintillation counters is in the field of γ -ray spectrometry, developed principally by P. R. Bell (ORNL). Some of the precautions that must be taken with, and the limitations of, the systems currently in use were discussed by R. K. Swank (Argonne) and W. H. Jordan (ORNL). Swank reported some observations on pulse height resolutions and photosensitivity of different multiplier tubes. He also described a spectrometer giving an energy resolution of 9%.

W. Jordan and his collaborators from the Oak Ridge National Laboratory showed scintillation spec-

trograms on which could be observed such details of structure as x-ray escape peaks, γ -ray back-scattering peaks, peaks due to x-rays generated in the surrounding shield, and peaks due to x-rays following internal conversion.

On Tuesday evening the guests of the symposium were invited to hear a talk given by L. Hafstad (NBS) on nuclear reactors. The entire symposium was characterized by a lively interest on the part of those attending, though it was unfortunate that the crowded program permitted little time for discussion during the sessions.

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Scientists in the News

Arthur F. Abt has accepted appointment as professor of pediatrics at the Duke University School of Medicine and director of the Radioisotope Unit for the Veterans Administration Hospital at Durham, N. C. Dr. Abt has been engaged in the practice of pediatrics in association with his father, Isaac A. Abt, since 1926 and has been active in public welfare organizations in Chicago.

Antonio Arena, former head of the Soils Division of the Argentine Ministry of Agriculture and later director of the Soils Institute, has been appointed soils specialist of the Technical Cooperation Program of the Inter-American Institute of Agricultural Sciences and will be stationed in the Southern Zone, with headquarters at Montevideo. **Luis Carlos Cruz Riascos** has been named executive secretary of the program and will function at Institute headquarters in Turrialba, Costa Rica. Mr. Cruz has been head of the Agronomic Service of the Chilean Nitrate Corporation in Colombia.

Alton E. Bailey, president of the American Oil Chemists' Society, has been chosen by the American Chemical Society's Georgia Section to receive the 1952 Herty Medal. The award, sponsored by the Chemistry Club of the Georgia State College for Women, is given annually for outstanding contributions to chemistry in the Southeast. Mr. Bailey, who is director of research of the Humko Company, Memphis, will receive the medal at the annual Herty Day celebration on the Milledgeville campus May 3. The Herty Medal is named in honor of the late Charles H. Herty, twice president of the American Chemical Society and originator of the process for manufacturing paper from southern pine.

David Birnbaum, who holds the Magnus-Warburg Fellowship from the Hadassah-Hebrew University Hospital in Jerusalem, is spending a year in New York City, working in the Gastroenterology Research Laboratory of the Mount Sinai Hospital under the direction of Franklin Hollander.

The vacancy on the Pacific Science Council created by the death of Sir Peter Buck has been filled by the Bernice P. Bishop Museum with the appointment of Cyril E. Pemberton, entomologist with the Experiment Station of the Hawaiian Sugar Planters' Association, Honolulu. Dr. Pemberton has been closely associated with the Pacific Science Association, having been chairman of the Standing Committee on Economic Entomology between the sixth and seventh Pacific Science congresses. He is now a member of the Standing Committee on Crop Improvement in the Pacific Area. Louis Malleret has been designated by the Academie des Sciences as council member from the Etats Associes de l'Indochine.

Viscount Chaplin has been appointed honorary secretary of the Zoological Society of London, succeeding Sheffield A. Neave, who is retiring after 10 years in office. Dr. Neave retired from the directorship of the Commonwealth Institute of Entomology in 1946.

Kingsley Davis, associate professor of sociology and director of population research at Columbia University, has been engaged by the Conservation Foundation to survey existing activities in various countries in regard to the relationship of natural resources and population pressures, under the terms of an anonymous grant of \$22,500 to the foundation.

Recent visitors at the National Bureau of Standards were Barend de Loor, of the Department of Statistics, University of Pretoria, South Africa; S. A. Sathar, deputy chief engineer, Posts, Telegraphs and Telephones, Karachi, Pakistan; and Bungo Umemiya, director and chief of the Research Department of the Japanese Spinners' Inspecting Foundation, Osaka. Professor de Loor is on a six-month visit in the U. S., where he is studying statistical research techniques and related matters.

Gaps in the history of the Roman occupation of England are being filled in by new discoveries and excavations, the most recent of which are at Puddle Hill, north of Dunstable, Bedfordshire, and on the site of Lloyd's new building on Lime Street, London. Excavation at Puddle Hill is being directed by Joan Evans, of the Luton Art School, and James F. Dyer, of the Luton Grammar School. The work at Lloyd's is being done by the Guildhall Museum staff under Norman Cook.

Ernest Carroll Faust, William Vincent professor of tropical diseases and hygiene, and head of the Division of Parasitology, Department of Tropical Medicine and Public Health, Tulane University, attended the first National Congress on Tropical Medicine, Lisbon, in April as Official Honor Guest from the U. S. This congress commemorated the 50th anniversary of the founding of the Institute of Tropical Medicine of Lisbon. Dr. Faust was asked to address the congress on "The Control of Schistosomiasis," "Clinical and Public Health Significance of Amebiasis," and "Some Morphologic Characters of *Diphyllobothrium latum*." Dr. Faust will also visit the De-

partment of Parasitology, School of Medicine in Paris, and the Institut Pasteur de Paris.

James J. Gibson, Cornell University, was awarded the Warren Medal by the Society of Experimental Psychologists at its annual meeting. The citation for the award was as follows: "The Howard Crosby Warren Medal is awarded to James J. Gibson for his many studies, culminating in his book, 'The Perception of the Visual World,' directed upon fundamental as well as very practical problems in the field of perception. His book is one of the few important works ever done on the central, neglected question as to how man comes to know the world about him."

Heinz E. R. Gruner has been appointed head of Bausch & Lomb Optical Company's Photogrammetric Section. A graduate of the University of Dresden, Dr. Gruner joined the firm's Scientific Bureau in 1948. During 1932-36 he was attached to Wright Field as civilian head of the Research and Development Group of the Army Corp of Engineers.

Carl G. Hartman, associate director of the Ortho Research Foundation, Raritan, N. J., has accepted an invitation to the faculty of the Free University of Berlin to give a course of lectures in physiology during the summer semester. He will also participate in the Ciba Foundation's Conference on Germ Cells and the annual meeting of the British Society for the Study of Fertility in London June 18-20 and June 25-27, respectively. He will return via Sweden after attending the II International Congress of Physiology and Pathology of Animal Reproduction in Copenhagen July 7-11.

John Jackson, who retired last July as His Majesty's Astronomer at the Royal Observatory, Cape of Good Hope, S. A., has been awarded the gold medal of the Royal Astronomical Society for 1951, in recognition of his work on stellar parallaxes and proper motions.

J. Elliott Janney, who is in charge of research and development for Rohrer, Hibler & Reople, of Cleveland, Ohio, will spend July in England interviewing a number of top British scientists in the course of an investigation of the process of creative thinking.

Z. Kopal, formerly of the Harvard Observatory, has recently been appointed professor of astronomy at the University of Manchester. His appointment strengthens the astrophysical trend in the university, which is already supporting the radio-astronomy program at Jodrell Bank under A. C. B. Lovell, investigations on geomagnetism, and several projects in theoretical astrophysics. An experimental astronomy program will begin shortly at Manchester, when the installation of an 18-inch reflector at Jodrell Bank is completed.

The Earl of Limerick, who was recently made a member of the Medical Research Council of Great Britain, has been elected chairman, succeeding the late Viscount Addison.

John Lounsbury, who has been working on the land utilization program in Puerto Rico, has recently been appointed to the staff at Antioch College, to teach geology and geography.

Andrew McCance has been appointed chairman of the Mechanical Engineering Research Board of the British Department of Scientific and Industrial Research, succeeding **Henry Guy**, who has just retired.

John P. McGovern, assistant professor of pediatrics, George Washington University, has received \$8000 in grants for research in the use of new drugs in the treatment of infants and children. A grant of \$3500 has been received from Charles Pfizer & Co. for studies with antibiotics, and White Laboratories has granted \$4500 for an evaluation of molybdenum in compounds containing iron to determine its effectiveness in the treatment of iron deficiency anemias in infants and children. Dr. McGovern joined the university staff in 1950 as a Markle Foundation scholar in medical science.

Frank A. Melton, professor of geology and aero-geology in the School of Geology, University of Oklahoma, received the Talbert Abrams Award for his paper "The Geomorphology and Photo-Geological Study of the Flat-Lands," published in *Photogrammetric Engineering* in December 1950.

The Cross of Alfonso X, "El Sabio," has been awarded to **José M. Otero** for his distinguished work in physiological optics and for his achievements in the organization and administration of science in Spain. Professor Otero is the director of the Instituto de Optica Daza de Valdes in Madrid. He is also director of the Spanish Naval Research Laboratory and vice president of the Commission on Nuclear Energy.

Oleg Polunin, science master at Charterhouse; **W. R. Sykes**, of the Royal Horticultural Society Gardens; and **L. H. J. Williams**, of the Department of Botany, British Museum (Natural History), are in Nepal on a botanical expedition sponsored by the museum and the society, and supported by other scientific organizations. The object of the expedition is to collect specimens of the flora native to the Himalayas from the Indian to the Tibetan boundaries, and from altitudes as low as 2000 feet and as high as 26,795 feet. The region is unexplored botanically.

R. Rivlin, of the British Rubber Producers Research Association and the Royal Institution (London), plans to spend six months in the U. S. this year. He will be at the Naval Research Laboratory as consultant in the Mechanics Division.

Melvin A. Schadewald has accepted appointment as assistant professor of pharmacology and toxicology at the University of Texas Medical Branch, Galveston. Dr. Schadewald has been research scientist at the Hastings State Hospital, Hastings, Minn.

E. W. R. Steacie, who has been vice president (Sci-

tific) since 1950 of the National Research Council of Canada, and director of NRC's Division of Chemistry since 1939, has been named president. He succeeds **C. J. Mackenzie**, who has been appointed president of the newly created crown company, Atomic Energy of Canada, Ltd.

The American Cancer Society has honored **Norman Treves**, founder and present director of the Nassau Tumor Clinic, with a medal and certificate. Dr. Treves is associate attending surgeon at Memorial Hospital for the Treatment of Cancer and Allied Diseases in New York City and also assistant professor of clinical surgery at Cornell University Medical School.

A. G. Walker, professor of mathematics at the University of Sheffield since 1947, has been appointed professor of pure mathematics at the University of Liverpool, succeeding **J. M. Whittaker**, who has been named vice chancellor of the University of Sheffield.

Grants and Fellowships

Allied Chemical & Dye Corporation will award 41 graduate fellowships (10 more than in 1951-52) for study in 26 universities and other academic institutions in the U. S. and Canada, principally in chemistry and chemical engineering. Each fellow will receive tuition in addition to a stipend of \$1500.

The **American Institute of Nutrition**, at its meeting in New York last April, presented the Borden Award in Nutrition (\$1000 and a gold medal) to Max Kleiber, of the University of California at Davis, for his contributions to knowledge of food utilization and energy metabolism of dairy animals; the Mead Johnson Award on the Vitamin B Complex (\$1000) to Howarde E. Sauberlich, of Alabama Polytechnic Institute, in recognition of his fundamental investigations of the citrovorum factor and its relation to folic acid; and the Osborne and Mendel Award of the Nutrition Foundation (\$1000) to Icie Macy Hoobler, of the Children's Fund of Michigan, for her extensive fundamental studies on the nutritive requirements of children.

National Medical Fellowships, Inc. (formerly Provident Medical Associates), has approved new fellowship and scholarship grants to 43 individuals for the year 1952-53. All awards are to Negro physicians and medical students, and they provide for training in dermatology, internal medicine, obstetrics and gynecology, ophthalmology, orthopedics, pediatrics, psychiatry, radiology, and surgery. The affairs of the organization are administered by an interracial Board of Directors, which has recently been increased from 9 to 15 members.

The **National Science Foundation** has approved an additional 41 research grants, totaling \$406,660, in the biological and physical sciences. Grants in this group range from one to five years. Largest grants went to the University of Missouri for research, under the direction of Arthur R. Laufer, on acoustic evita-

tion (\$31,700); to Purdue, for an investigation under Herbert C. Brown of the effect of structure on chemical reactivity using molecular addition compounds (\$25,300); and to Chicago Natural History Museum, for a taxonomic study of the tropical plants of Colombia by José Cuatrecasas (\$25,000). Funds were distributed as follows: biochemistry, 2 grants; biophysics, 4; chemistry, 11; comparative physiology, 1; developmental biology, 1; endocrinology, 1; engineering, 1; enzyme chemistry, 1; experimental plant biology, 2; genetics, 2; microbiology, 2; physics, 5; pharmacology, 2; research education in the sciences, 1; systematic biology, 5.

Meetings and Elections

The American Gas Association has elected Charles E. Bennett president to succeed the late George F. Mitchell. Mr. Bennett is president of the Manufacturers Light and Heat Company, Pittsburgh. Frank C. Smith, of Houston Natural Gas Company, was elected vice president.

At its annual meeting in March in Salt Lake City, the American Mosquito Control Association elected C. R. Twinn president to succeed Don M. Rees. Dr. Twinn is head of the Veterinary and Medical Unit of the Canadian Department of Agriculture.

The American Psychosomatic Society has elected the following officers: president, Sydney G. Margolin; president-elect, George L. Engel; secretary-treasurer, Fredrick C. Redlich. David T. Graham, George C. Ham, Erich Lindemann, and Milton Rosenbaum were elected to the Council.

An Association of Indian Science Writers was organized during the Indian Science Congress recently held in Calcutta. D. M. Bose, one of the editors of *Science and Culture*, was appointed chairman of a committee of 19 to draft a constitution for the new society. S. N. Sen was made secretary of the committee.

The first Plansee Seminar "De re metallica" will be held June 22-26 in Reutte in the Tyrol, under the auspices of the U. S. High Commissioner for Austria, the Austrian minister of education, the rector of the University of Innsbruck, and representatives of the Tyrolean government. The tentative program includes lectures by these American scientists: G. J. Comstock, V. C. Frechette, H. H. Hausner, W. J. Kroll, J. T. Norton, and J. Wulff.

The Institute of Metals has elected the following officers: president, C. J. Smithells, of British Aluminum Company; vice presidents, G. L. Bailey and S. F. Dorey; treasurer, E. H. Jones.

A Symposium on Electron Transfer and Isotopic Reactions will be held at the University of Notre Dame June 11-13, under the joint auspices of the Division of Physical and Inorganic Chemistry of the American Chemical Society and the Division of Chemical Physics of the American Physical Society.

Miscellaneous

The American Museum of Natural History, the National Geographic Society, and Dr. and Mrs. Carnes Weeks, of Medford Plantation, Mount Holly, S. C., are sponsors of an expedition to French Equatorial Africa and the French Cameroons for a study of the little-known flora and fauna of those sections. Dr. and Mrs. Weeks; Walter Weber and Volkman Wentzel, of the National Geographic; Donald Carter, of the American Museum; and Ernst A. Zwilling and Kurt Uetz, of the Haus der Natur Museum, Vienna, will be members of the expedition. Extensive sound recordings of native dialects and music will be made.

National headquarters of the American Rose Society will be moved from Harrisburg, Pa., to Columbus, Ohio. The city has voted to spend \$205,000 to establish the Park of Roses, "the largest rose garden in the world." It will contain about 50,000 rosebushes.

The Commission on Chronic Illness will move to Baltimore July 1 for a special, long-term, intensive study of chronic disease among 4000 Baltimore families, a companion study to one being made on the rural population of Hunterdon County, New Jersey. Dean W. Roberts, of Johns Hopkins, deputy director of the Maryland State Department of Health, is the newly appointed head of the commission.

Chemicals wanted by the Registry of Rare Chemicals, 35 W. 33rd St., Chicago 16, include: calcium palmityl phosphate; methyl aluminum diechloride; ruthenium fluoride; sodium hexavanadate; triphenyl antimony; 2,5-xylyl hydrazine; piperonylic acid; purpurin sulfonate; mesoxalic acid; 1-hydroxymethylcyclobutan-1; decamethylene diamine; divinyl sulfone; choline stearate; 0,3,3-bicyclooctane; areeinidine; arginase; coronene; eupreine; luciferin; dihydroquercetin; and o-tert-butyl toluene.

Soviet physicists won top honors in the annual award of Stalin Prizes for outstanding scientific work. Nikolai Vasilevich Belov, the late Sergei Vavilov, Elphidizer Kirilov, Vadim Levshin, and Eleuter Andronikashvili were the physicists honored; other prizes went to Vladimir Nagovsky and three co-workers of the Academy of Medical Sciences and to Anatolii Tomashevich for work in the military sciences.

Vol. I, No. 1, of the following have been received: *Anais de Microbiologia*, University of Brazil; *The Archives of Orthodontics*, bimonthly, \$12.00 per year (Lucien DeCoster, editor-in-chief); *Science of Light*, from the Institute for Optical Research, Tokyo University of Education (in Japanese with English abstracts—succeeding volumes to be in English). *Orthopedic and Prosthetic Appliance Journal*, a quarterly, succeeds the *Journal of the Orthopedic Appliance & Limb Manufacturers Association*; and the *Nagoya Journal of Medical Science*, of Nagoya University, Japan, has resumed publication, after 11 years, with Vol. 14, No. 1.

Technical Papers

The Separation of Porphobilinogen and an Ehrlich Negative Precursor of Uroporphyrin¹

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Hitherto it has been widely accepted that the Ehrlich aldehyde reacting porphobilinogen, excreted in the urine of acute porphyria cases, is the precursor of both uroporphyrin and the nonporphyrin pigment, porphobilin. Waldenström and Vahlquist (1) reported that the heating of porphobilinogen-containing urines at pH 4.0 favored uroporphyrin formation, whereas heating at pH 8.0 resulted in a greater formation of porphobilin. They observed the same behavior with porphobilinogen solutions obtained by electrophoresis and regarded as homogeneous entities. Evidence has now been obtained, however, that the Ehrlich-reacting porphobilinogen gives rise only to porphobilin once it has been separated from one or more non-Ehrlich-reacting precursors, which regularly accompany it in the urine of these cases. On the other hand, it has become clear that variable, but often large, fractions of the uroporphyrin in these urines are obtained from non-Ehrlich-reacting precursors. A relatively simple method of separating these substances has now been worked out and is described briefly here.

Urine samples from 8 patients suffering with intermittent acute porphyria were filtered and acidified to pH 5.6-6.0 with acetic acid. A column of Merck's alumina (for chromatographic analysis) was washed with 1% acetic acid until the eluate was acid to litmus. The filtered urine was then run through with gentle suction. Both pigments and chromogens were adsorbed on the column, which was washed first with distilled water and then with 1% acetic acid. The latter contains the porphobilinogen in a relatively concentrated solution. In some cases small amounts of uroporphyrin precursor are also eluted. This solution was brought to pH 6.0 and then irradiated in a Pyrex test tube for 3-5 min with filtered light from an ultraviolet lamp.² The irradiation converts any uroporphyrin precursor that may be present to porphyrin, leaving the porphobilinogen unaffected. The solution was barely alkalized with dilute NaOH, and the free porphyrins were precipitated with CaCl_2 and Na_2HPO_4 , according to the method of Sveinsson, Rimington, and Barnes (2). This final Ehrlich-reacting filtrate was usually free of porphy-

¹ This study was supported in part by a grant from the U. S. Public Health Service (RG 345) and under contracts with the Atomic Energy Commission, #At (11-1)-100, and the Surgeon General's Office, U. S. Army, under sponsorship of the Commission on Liver Disease, Armed Forces Epidemiological Board.

² Model BL-2, Black Light Products, Chicago, Ill.

rin and porphyrin precursors. In certain instances it was necessary to repeat the radiation and CaHPO_4 adsorption of uroporphyrin in order to separate the porphobilinogen completely from uroporphyrin precursor.

Solutions of porphobilinogen obtained by the above method, after having been acidified to pH 4.0 and boiled for 30 min, gave rise only to porphobilin, a dark-brown pigment without characteristic absorption spectrum or any porphyrin characteristics.

Electrophoretic studies to be described in detail separately have revealed that porphobilinogen and the uroporphyrin precursor have a single homogeneous zone and cannot be separated on the basis of electrophoretic mobility. The solution of the single zone, however, can then be subjected to the above-described method with complete separation of the two chromogens.

The present observations are believed to explain an incorrect concept that porphobilinogen is a precursor of uroporphyrin as well as porphobilin. Further studies of these precursors are in progress.

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Ultraviolet Absorption Spectra as a Tool for Diagnosing Plant Virus Diseases^{1,2}

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and Bureau of Plant Industry, Soils,
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U. S. Department of Agriculture

Methods developed in this laboratory for the detection of viruses in fruit trees by means of color or staining tests (1-4), although useful, are limited in that they are not critical in separating one virus from another. Moreover, these methods are not applicable to herbaceous plants. In a search for more refined methods of virus detection, the ultraviolet absorption spectra of various extracts from virus-diseased and healthy leaves were investigated. After considerable study it was found that acid hydrolysis of leaf tissue, subsequent to the removal of the alcohol-soluble constituents, produced absorption

¹ Scientific Paper No. 1090, Washington Agricultural Experiment Stations, Pullman, Project No. 865.

² This investigation was supported in part by funds provided for biological and medical research by the State of Washington Initiative Measure No. 171.

spectra that appeared to be characteristic of the virus diseases studied. More than 3000 healthy and diseased plants of various species have been studied by this procedure, which shows promise of being helpful in the study of virus diseases. Much of the work has been confined to virus diseases of stone fruits, but the method is suitable for virus diseases of herbaceous plants as well.

A relatively small sample is required for the procedure. Two leaf disks, $\frac{1}{4}$ " in diameter, are taken with a paper punch. Usually each disk is taken across the mid-vein or a large lateral vein to insure the inclusion of both phloem and xylem tissues. More than two leaf disks may be required for certain types of plants or for certain diseases. In some cases, one disk may be enough. Very young leaves or very old ones should not be sampled; the sampling should be confined to mature leaves. For small plants, a minimum of three samples of two disks each per plant is used from leaves of different ages. It is desirable to test leaves of different ages because the virus concentration may vary considerably from the apical to the basal portion of any current growth. Moreover, if virus mixtures are present, the relative proportions of the constituent viruses may vary with leaf age. It is easier to detect the constituent viruses if leaves of different ages are sampled. The three samples from any given plant are analyzed separately, and the absorption curves are plotted separately. Fresh material has been used in this work to avoid possible loss of labile constituents that might occur in dried material.

The two leaf disks are placed in a test tube, 1 ml 70% ethyl alcohol is added, and the tube is heated in a water bath at 80° C for 10 min. The alcohol is decanted from the sample, a fresh 1-ml portion is added, and the extraction procedure is repeated. Three such extractions are usually sufficient to remove all alcohol-soluble material. The leaf disks should be free of chlorophyll at the end of the extraction period. The final alcohol wash is drained from the sample, and 3 ml of an alcohol-hydrochloric acid mixture is added (10 ml HCl plus 90 ml 95% ethyl alcohol).³ The sample is then heated at 80° C in a water bath for 30 min and cooled, and the alcohol-HCl mixture is decanted into a silica absorption cell (10 mm light path). A Beckman DU spectrophotometer was used in these studies, and absorption readings were taken at 2-m μ intervals in the range from 230 to 300 m μ . The alcohol-HCl reagent was used as a reference sample.

³ Alcoholic HCl was used for hydrolysis rather than aqueous HCl because with virus-diseased stone fruit leaves a red color is produced by alcoholic HCl, and thus a simple color test as well as the ultraviolet absorption test is available from the same sample. The intensity of red color may be measured independently in a photoelectric colorimeter prior to the ultraviolet absorption measurements. This color test is based on the same principle as the tests reported previously (1-4). In this particular color test the HCl presumably produces furfuraldehyde from pentose sugars, and the furfuraldehyde condenses with phenolic compounds and yields the red color. Similar reactions have been used as color tests for pentose sugars (5).

In tests of leaf tissue that we consider to be healthy, whether from a herbaceous plant or a fruit tree, characteristic absorption curves for nucleic acid or its hydrolysis products are obtained (Fig. 1). The

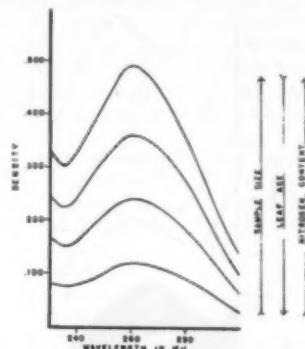


FIG. 1. Absorption curves from healthy plants, showing effect of sample size, leaf age, and nitrogen content.

height of the absorption peak at 260 m μ is a measure of the amount of nucleic acid present. The height of the peak may be increased by increasing the sample size or by using younger leaf tissue. Thus in comparing diseased tissue with healthy tissue it is necessary to use comparable samples.

The mosaic-type virus diseases that we have studied (tobacco mosaic and potato X) are not readily distinguished by this method because the principal hydrolysis products are nucleic acids (Fig. 2). Ring-

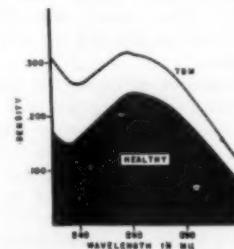


FIG. 2. Absorption curve from healthy *Physalis floridana* Rydberg compared with that of *P. floridana* affected with tobacco mosaic (TBM).

spot-type viruses, as represented by ringspot of stone fruit trees, produce a distinctive absorption curve, with a peak at or near 270 m μ (Fig. 3 A). The yellows-type virus diseases that we have studied, such as sour cherry yellows and Western X disease, also produce distinctive absorption curves, with peaks at or near 280 m μ , which appear to be characteristic (Fig. 3 B, C, D). Sour cherry yellows is considered to be caused by a complex of at least two viruses, ringspot virus and an unknown (6); this is confirmed by the absorption curves. The 270 and 280 m μ peaks are consistent for this disease, but the relative heights of the two peaks vary independently with a number of conditions (Fig. 3 B and C). Sometimes the ring-

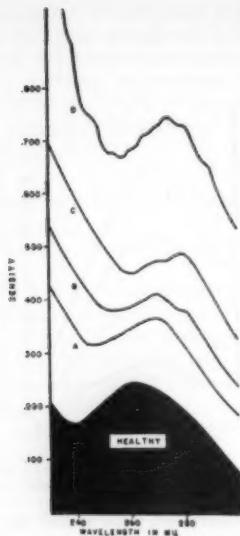


FIG. 3. Absorption curves from peach affected with various virus diseases compared with absorption curve from healthy peach: A, Ringspot; B and C, sour cherry yellows; D, Western X disease.

spot peak is dominant, and at other times the yellows peak is dominant.

Western X disease of peaches gives a very complex absorption curve (Fig. 3 D), which may be due to a mixture of several viruses. This disease from different sources often shows different secondary peaks, but the peaks at 274-276 μ and at 280-282 μ are consistent.

With the information on hand, it is not possible to determine whether each absorption peak is due to a single virus or whether a single virus may give more than one peak. If it were possible to study pure preparations of each of the viruses, this point could readily be settled; but so far none of the stone fruit viruses has been isolated in reasonably pure form. Likewise, it is not known whether the absorption peaks actually indicate hydrolysis products of the virus itself or end products of an interaction between the virus and the host. With a readily purified virus like tobacco mosaic, it is possible to show that the absorption peak obtained is probably from the virus itself. Fig. 4 shows the absorption curve of a purified preparation of tobacco mosaic virus along with the absorption curve of the hydrolysis products of the same preparation. A purified preparation of tobacco mosaic virus yields a nucleic acid absorption curve on hydrolysis that is similar to the curve obtained from leaves infected with the virus (*cf.* Fig. 4 and Fig. 2).

It has not been possible to determine whether disorders other than those caused by viruses produce materials in leaves that would interfere with this method. Nitrogen deficiency has been studied under

controlled conditions and found to affect merely the height of the normal nucleic acid peak. The higher the nitrogen content of the leaf, the higher the nucleic acid peak (Fig. 1).

In some types of plant material in which virus mixtures occur, the concentration of one of the components may be very high and mask the absorption curves of other viruses that may be present. This is often true of sweet cherries, in which the 280 μ yellows peak is very high.

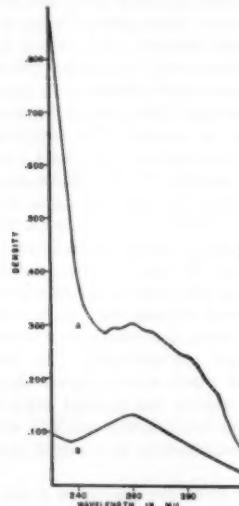


FIG. 4. Absorption curves of tobacco mosaic virus. A, 100 ppm purified tobacco mosaic virus in M/10 phosphate buffer at pH 7.0; B, alcoholic-HCl hydrolysis products of same preparation.

It should be emphasized that, as with all analytical methods, this one indicates only what is present at the time and place of sampling. Moreover, the virus concentrations must be high enough to produce an absorption curve that is distinctive from the curve from healthy tissue. Virus infection may be detected with this procedure before symptoms are obvious. For example, a stone fruit virus transmissible to cucumber usually produces visible symptoms in cucumber cotyledons in 4-5 days. With the spectrophotometer the disease can be detected in cucumber cotyledons 48 hr after inoculation.

Care must be exercised in associating any given symptom of a virus disease with a particular absorption peak if virus mixtures are present in a sample.

Although the method requires the use of a good spectrophotometer and is somewhat time-consuming (analyses of about 30 samples a day exclusive of plotting the absorption curves being possible), it promises to be very helpful in the diagnosis and study of virus diseases. It shows particular promise in the study of latent viruses, and virus mixtures and

complexes of stone fruits, and possibly may aid in arriving at a more rational virus nomenclature based upon chemical properties.

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Formation of 2,3-Butylene Glycol in Bacterial Fermentation of D-Glucosamine¹

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There have been a few reports made with respect to the products of bacterial fermentation of D-glucosamine. According to these papers, acetic and butyric acids (1), propionic and D-lactic acids (2), D-lactic, L-lactic, and succinic acids (3), etc., have been shown to be produced by action of bacteria on D-glucosamine.

In our laboratory extensive investigations were recently carried out on the ability of microorganisms in the soil to make use of D-glucosamine; those that were observed to utilize it were isolated into pure cultures. From among them, a number of active strains of bacteria and of fungi were selected for studies on the fermentation of D-glucosamine. The bacteria of the *coli-aerogenes* group were found to utilize it vigorously.

A report is made here on the products of the fermentation of D-glucosamine by *Aerobacter cloacae* (Jordan) Bergey *et al.*, isolated from soil. A liquid culture medium containing D-glucosamine alone as the source of both carbon and nitrogen was employed; composition of the medium was as follows:

(A) D-glucosamine hydrochloride	40.0 g	in 1 liter of water
(B) NaCl	10.0 g, KH ₂ PO ₄	4.0 g } in 1 liter
	MgSO ₄ · 7H ₂ O	0.2 g, CaCO ₃ 10.0 g } of water

where (A) and (B) were prepared apart, sterilized separately, and then combined aseptically.

A strain of *A. cloacae* was inoculated into the medium and incubated at 37° C for about 10 days. In the early stages of incubation, a vigorous generation of gas was observed.

After the incubation period, the liquid culture was filtered, concentrated *in vacuo* to about 300 ml, and

¹ Previously reported at the meeting of the Chemical Society of Japan in April 1947.

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³ The authors wish to express sincere thanks to Kendo Saito and Shiro Akabori for their kind guidance.

extracted continuously with ether for 50 hr. The ether extract was dried, concentrated to a viscous liquid, and the residual liquid was distilled under diminished pressure to give 6.9 g (approx 20% of the D-glucosamine) of the main fraction at 89°–92° C/16–17 mm Hg after a small quantity of the foregoing fraction (0.5 g). The substance obtained was a colorless, clear, viscous liquid ($[\alpha]_D^{25} = +1.3^\circ$ [1 = 1dm]), somewhat like glycerol, which crystallized in the cold. We confirmed it to be 2,3-butyleneglycol by distilling it with 25% sulfuric acid, followed by separating the methylglycidyl ketone, which is to be derived from 2,3-butyleneglycol, from the distillate, converting the ketone into the *p*-nitro-phenylhydrazine (yellow; mp, 126.5°–127.5° C), adopted under the Akabori method (4).

After the extraction of the glycol, the aqueous mother liquor was strongly acidified with sulfuric acid and extracted again with ether continuously for 40 hr. On concentration of the extract, there remained a considerable amount of colorless, clear, viscous liquid containing the crystals of succinic acid (1.1 g; approx 3% of the D-glucosamine) and giving out an acetic acid smell. The volatile acids contained were separated from it by steam distillation as usual; the total amount was estimated at 0.64 g as acetic acid. Lactic acid was also found in this acidic extract.

In addition, small amounts of ethyl alcohol and acids fixed in the precipitate, such as oxalic acid, were also observed to be produced in this fermentation.

Study as to when and how the amino group is split off in the process of the fermentation of D-glucosamine is in progress.

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The Influence of Skin Temperature upon the Pain Threshold as Evoked by Thermal Radiation—A Confirmation

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In a recent paper Hardy, Goodell, and Wolff (1) reported experiments on the effect of skin temperature on pain threshold evoked by thermal radiation. By a graphical extrapolation from their data, they were able to infer that "the skin in the areas tested must be raised [to a temperature of 44.9° C] to be noxiously stimulated, regardless of the initial level of skin temperature."

¹ This research was carried out while the author was at the Psycho-Acoustic Laboratory, Harvard University, Cambridge, Mass.

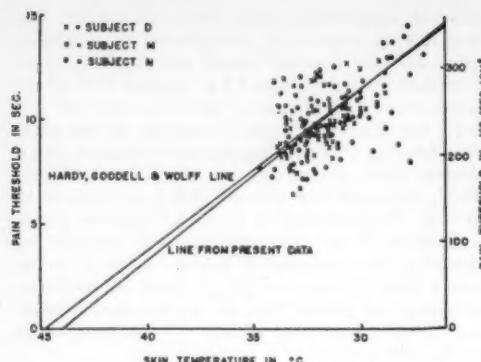


FIG. 1.

The present report derives from an investigation of the temporal variation of various sensory thresholds, in which skin temperature was measured as a control for pain threshold. The resultant data are comparable to those of Hardy, Goodell, and Wolff, except that the range of the present data is smaller than theirs, because no attempt was made to produce major changes in skin temperature artificially.

At 7 p. m. daily for 45 consecutive days pain thresholds were measured on each of three subjects. Measurement was performed on an apparatus designed after that of Hardy *et al.*, but with a different technique.

The radiant intensity was set at 275 mc/sec/cm² each time and was applied to the forearm on a blackened circular area 1.8 cm in diameter. The threshold was measured in terms of the number of seconds that the radiation had to be applied before the subject reported burning pain.

Just before application of the stimulus, skin temperature at the blackened spot was taken by means of a thermistor (1 V6 11) and a GR type 150 impedance bridge; resistance was translated into degrees centigrade by means of a calibration curve.

Correlation coefficients between threshold and temperature were -0.70 , -0.61 , and -0.72 for the three subjects, and all were highly significant statistically (better than the 1% level). The data were plotted (Fig. 1), and a least square straight line was fitted to the data in a manner comparable to that in Fig. 1 of Hardy, Goodell, and Wolff. Extrapolation from this line provides a striking confirmation of their "zero point," since our data yield a value of 44.1°C in comparison with their report of 44.9°C . The agreement is all the more remarkable because Hardy, Goodell, and Wolff—

- 1) Used threshold for pricking pain, whereas we used that for burning pain;
- 2) Used the Hardy-Wolff-Goodell method, and we used a very simple temporal measure;
- 3) Used a temperature range of over 20°C , whereas we used one less than half as large; and the variability of our data was even larger than theirs.

The present experiments provide further confirmation of Hardy, Goodell, and Wolff's conclusion:

Buettner and Heniques and Moritz have shown that reversible tissue damage in the skin of the forearm and upper leg of humans is produced at the critical temperatures of 44°C to 45°C . In the above experiments threshold pain has been elicited when the skin temperature has been raised to roughly this same level, irrespective of the initial level of skin temperature. From these two independent observations the close relation between tissue damage and noxious stimulation can be inferred, thus significantly supporting the concept that the adequate stimulus for pain is tissue injury.

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Time Distortion in Hypnosis and Nonmotor Learning

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By "time distortion" is meant a marked difference between the seeming duration of a time interval and its actual duration, as measured by the clock. In previous communications on this subject (1, 2), evidence was presented which indicated that the following statements are probably true:

- 1) In especially trained subjects, time sense can be deliberately altered to a predetermined degree by hypnotic suggestion. Such subjects can have an amount of subjective experience under these conditions that is more nearly commensurate with the subjective time involved than with the world time. This activity, although seeming to proceed at a natural rate as far as the subject is concerned, actually takes place with great rapidity relative to world time.
- 2) Retrospect falsification or elaboration does not enter into the subject's reports.
- 3) The continuity of these experiences during distorted time is good.
- 4) Thought, under time distortion, although apparently proceeding at a normal rate from the subject's point of view, can take place with extreme rapidity relative to world time. Such thought may be superior, in certain respects, to waking thought.

Thus, apparently, time can be given to the hypnotized subject, and he can use this time for various mental activities.

It is important for investigators to realize that the training of a subject for time distortion generally has required 3-20 hr, exclusive of his training as a hypnotic subject *per se*. Training was generally carried out on consecutive days, the average session lasting one hour. With sufficient effort and the proper technique, the phenomenon can probably be produced to varying degrees in the majority of hypnotic subjects.

In discussing time distortion in hypnosis, world time is solar, or clock, time, and personal time is sub-

jective, or experiential, time. Estimated personal time is the estimate, by the subject, of an interval of his experiential time. Suggested personal time is a time interval that is suggested to the subject under hypnosis. Allotted time is the time, in world time, that is allotted to a task by the experimenter. A completed activity is one that progresses to the fulfillment of certain stipulated or implied conditions (none of them concerning time), at which point it reaches completion. An incomplete or continuous activity is one that does not progress to such a limit. Distorted subjective time during hallucinated activity as experienced by subjects in these and similar experiments is sometimes referred to as special time, or special trance time.

In this paper, presentation time is the time in seconds required to present verbally to the subject the material to be learned. The term "study period" connotes the time allowed the subject for the study of the material presented, and thus does not include presentation time. Its onset was indicated by the signal "Now," or "Take."

The purpose of this study was to compare two methods of learning nonsense material. In one, the subject employed certain learning techniques while awake; in the other, he employed the same techniques in the hallucinated world, under conditions of time distortion, while in the trance state.

The subject was a 22-year-old, single, male graduate student in psychology. He was cooperative and intelligent and an excellent hypnotic subject. He had had considerable experience with the experimental use of nonsense syllables, and had been trained in time distortion. Two series of 150 paired nonsense letter-groups of 3 letters each were used. Some of these were syllables; others were consonant groups. Each pair was printed with blue crayon, in large type, on a $3'' \times 5''$ card thus: CGJ—QXH. The series were of comparable difficulty.

The task was to learn to give correctly, within 3 see, the second group in the pair, in response to the verbal presentation of the first group by the experimenter. At each daily session, 5 pairs from one series of letter-groups were learned by the subject while awake, and 5 pairs from the other series while he was in the trance state. Twenty-four hr later he was tested for retention and relearning. Waking and trance tasks were assigned in alternate order on successive days.

Learning. At each session there was an initial basic study period, followed by successive runs through the 5 cards.

1. AWAKE

a) Basic study period: The subject sat at a table with pencil and paper at hand, and was instructed to print out each letter-group-pair 5 times, saying them over to himself and forming associations while doing so. He was to start printing immediately after the presentation of a given pair. The experimenter, in a period of 10 sec, then read the first pair of letter-groups to him thus: "CGJ, dash, QXH—CGJ, dash,

QXH—Now." The "Now" marked the end of the 10 sec, and was the starting signal for the subject to print the material 5 times. As he printed, he said the letters to himself and tried to form associations. When this was finished, the pair on the next card was presented in the same way. The average time taken by the printing was 26.5 sec, making a total average time of 36.5 sec.

b) Runs: On completion of the above, the runs were begun, each preceded by a shuffling of the cards. The subject sat with his eyes closed. The experimenter read aloud the first group of one of the pairs, and the subject immediately stated the second group if he could. At the end of 3 sec, the experimenter said, "Take," whereupon the subject opened his eyes and looked at the card. Then followed a 5-sec study period, during which he repeated the letters, and formed associations, but did no printing. This period was terminated by the presentation of the first letter-group of the next pair. Subsequent runs followed immediately and continued until all the responses were correct. The maximum number of runs required was four.

It is thus evident that a run constituted both a test of performance and an opportunity for learning or for reinforcement.

2. TRANCE STATE

A moderately deep trance state was induced by suggestions of sleep.

It is important to point out that the following suggestions and instructions pertain to the subject's experience in the hallucinated world only. This world, incidentally, is very real to him, and it is in this, rather than in the physical one, that he carried out his trance study in distorted time, including the (purely hallucinated) printing. Throughout, he remained motionless, with his eyes closed.

a) Basic study period: The following suggestions were given to the subject:

You're now going to learn some nonsense letter-groups. You will have ample time between signals to learn them solidly. [This reference to "ample time" was understood by the trained subject to refer to his "special time."] As I give you a pair, you will print it out five times, exactly as you did while awake, saying the letters to yourself and forming associations as you do so. After that you may print them some more, or say the letters over to yourself many times more in order to take advantage of repetition, or form unusual associations, or adopt any other method of learning that you wish. This activity will impress the material upon your memory. It is important that you take as much of your special trance time as is necessary. You will not hurry, and it will be easy to learn them and to recall them later.

During the last 13 runs the above suggestions were given verbatim. Prior to that, the same ideas were presented, but the wording was varied slightly at times.

Immediately after giving the suggestions, the actual presentation of the material was begun. The subject sat at a table with his eyes closed, and the letter-

groups were read to him exactly as in the waking state. Then, at the starting signal, he began his trance study, using distorted time. At the end of only 5 sec allotted time, the amount being unknown to the subject, the experimenter gave the termination signal ("Now, blank"), indicating to the subject that he was to stop all mental activity and make his mind a blank. The next pair was then presented. Thus the presentation time was 10 sec, and the hallucinated activity lasted 5 sec.

b) Runs: During the runs, the technique was identical with that used in the waking state, except that the subject, in the 5-sec study period, closed his eyes again and, using distorted time, practiced much as he had during the basic study period for the trance state.

Retention test and relearning. Here the subject was awake throughout. There was of course no basic study period. Runs were assigned until the responses were all correct. The technique was identical with that already described for runs in the waking state. This testing was generally done 24 hr after the original learning, but in some instances several days intervened.

Tables 1-3 show the subject's performance.

TABLE 1
PROGRESS IN LEARNING

Study period	Waking				Trance state			
	I* (Sec)	II† (Pairs)	III‡ (Pairs)	IV§ (%)	I* (Sec)	II† (Pairs)	III‡ (Pairs)	IV§ (%)
Basic	26.5	150	62	41.3	5	150	89	59.3
First run	5	88	54	61.4	5	61	51	83.6
Second run	5	34	25	73.5	5	10	8	80
Third run	5	9	5	55.5	5	2	2	100
Fourth run	5	4	4	100				

* Av duration of study period.

† Total number of unlearned pairs studied during the 30 study periods of a given designation.

‡ Total number of pairs learned in the 30 periods of a given designation.

§ Percentage learned of the total unlearned pairs presented during the 30 periods of a given designation.

TABLE 2
AVERAGE LEARNING TIME

Av learning time per letter-group	Waking (sec)	Trance state (sec)
Including presentation time	41.0	17.4
Not including presentation time	31.0	7.4

TABLE 3
RETENTION AND RELEARNING

	After waking learning	After trance learning
Retention	24%	28%
Av relearning time per letter-group-pair	7.6 sec	6.1 sec

The subject stated that trance study in distorted time seemed easier than waking study. Not only did he have more time for associations, which in addition came more easily, but he had the benefit of rote practice. He always had plenty of time, and felt that he had really learned the material by the end of the study period, even if he didn't always remember it. He did not hurry, and all activity seemed to him to proceed at the normal or customary rate. Although each study period lasted only 5 sec, it seemed to him to be 4 or 5 min.

Those interested in these findings should read the previous reports (1,2) for an understanding of time distortion in hypnosis and its implications. It should be mentioned here, however, that the tasks involving trance study, as described above, fall into the category of completed activities, since they proceed to a specified point—i.e., the learning of the letter-groups. The allotted time was 5 sec. No specific interval of suggested personal time was used, but the subject was told that he would have all the time he needed. The estimated personal time was 4 or 5 min.

The average learning time per letter-group-pair, including the time required for presentation of the material, was 41.0 sec in the waking series, and 17.4 in the trance series. If the presentation time of 10 sec is not included, the times are 31.0 sec and 7.4 sec, respectively.

The results certainly indicate that the study in the trance state in distorted time was more effective than that in the waking state. Further investigation will be necessary to determine the relative significance, in this apparent facilitation of the learning process, of such factors as increased motivation and better concentration on the one hand, and of true "utilization" of special trance time on the other. We believe that the latter is of considerable importance and that the allotted time could probably have been cut down from 5 to 3 sec or less without materially altering the results.

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Manuscript received December 13, 1951.

Comments and Communications

Use of Nembutal as an Anesthetic for Large Wild Mammals¹

DURING the study of hibernation in the black bear, it was necessary to attach instruments to the animal and to take periodic samples of blood. This required the complete immobility of the bear. Ether anesthesia took too long a time before complete sedation occurred. Nembutal (the trade name of sodium pentobarbital), because of its ease of administration, safety, and rapid action, was therefore tried and found satisfactory.

An adult male American black bear (*Ursus americanus*), weighing 211 pounds, was made available through the courtesy of Edward Johnson, superintendent of the Woodland Park Zoological Gardens, Seattle, Wash., and was anesthetized upon three occasions. Several attempts at oral administration of Nembutal, both in a bolus of ground raw meat and mixed with powdered brown sugar, proved unsuccessful, so intraperitoneal injections were used. The bear, without food for 24 hr, was incarcerated in a squeeze cage and held steady while the Nembutal, dissolved in water at a concentration of 25 mg/kg body weight, was injected intraperitoneally. The bear was then allowed to move freely within the cage, while observations and records were made on its heartbeat, respiration rate, and subsequent actions until sedation occurred.

Immediately following the injection a respiration rate of 28/min and pulse rate of approximately 160/min were recorded. Within 17 min surgical sedation was attained. The respiration rate dropped and remained fairly uniform at 12/min, with the pulse rate at approximately 110. Surgical sedation on this occasion remained for about 83 min, during which many blood samples were taken from the saphenous vein. The bear showed no deleterious aftereffects. The data obtained from these and subsequent blood samples are being described elsewhere.

A second intraperitoneal injection with a smaller dose of 20 mg/kg body weight proved inadequate, so a supplementary injection of sodium pentothal, 7.5 mg/kg, was given intraperitoneally. In approximately 43 min after this second injection, or 2 hr and 5 min after the initial injection of Nembutal, sedation occurred. Food had not been withheld for the previous 24 hr, and this might have prolonged the time required. Although sedation was not as deep as in the previous trial, since both the heartbeat at 120/min and the respiration rate at 22 were higher, nevertheless, it persisted for approximately 1 hr. No ill aftereffects were noticed.

A third trial similar to the first was made after food had been withheld for 24 hr. In 13 min complete

¹ Acknowledgment is made to the Air Force under contract AF 33 (088)-18509 for aid and assistance in making this study.

surgical sedation, which lasted approximately 1 hr was reached, with the pulse rate remaining at about 100/min and a respiration rate of 12. Blood samples were taken, and electrocardiograms, involving numerous manual operations in the cage with the bear, were recorded. Again there were no ill aftereffects. Repetition of dosage and effects indicated that Nembutal injected intraperitoneally at a concentration of 25 mg/kg body weight can be safely administered to bears.

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The Peyote Cult

I WISH to express appreciation for, and to concur with, the communication dealing with peyote, signed by Messrs. La Barre, McAllester, Slotkin, Stewart, and Tax, in your November 30 issue. The subject is of importance to American Indians, to civil liberties, and to anthropology.

In late 1922, when federal and state proscriptions against the Native American (Peyote) Church were being pressed, the peyote cult members at Taos Pueblo laid their case before me. They offered (with an understanding of what was involved) to submit themselves individually and as a group to the fullest scientific investigation. They understood that such investigation would be pharmacological, biological, psychological, and social, and that it would involve experimentation, using part of their number as a control group.

In 1924, at the meeting of the Committee of 100 on Indian Problems (a Committee assembled by Secretary of the Interior Hubert Work), I reported this offer; a resolution was enacted, calling upon the National Research Council to plan and execute an investigation into peyote. The council never initiated this requested investigation.

Some years later, for the American Indian Defense Association, Donald Collier (at present a staff member of the Chicago Natural History Museum) canvassed all the then-existing literature on peyote, totaling some 400 published books and papers. His conclusion was identical with that set forth in the communication in SCIENCE.

Subsequently, in 1933, I became U. S. Commissioner of Indian Affairs. I introduced the above-mentioned analysis of the existing literature into the *Congressional Record* (the hearings, as I remember, of the House Sub-Committee on Interior Department Appropriations); and Secretary Harold L. Ickes and I prohibited absolutely any interference by the Indian Bureau with the religious practices of the Native American Church. We were abused in a good many quarters for this action; but the administrative

policy that we established has been adhered to up to the present time, although the Navajo tribe has passed and tried to enforce ordinances against the use of peyote. Administratively, we felt that we had no right to veto the Navajo Tribal Council's action.

There remain on the books of a number of states (procured by the Indian Bureau and by missionaries in earlier times) statutes that declare the ceremonies of the Native American Church (in effect) to be misdemeanors; and there is an occasionally renewed drive at Washington to secure the classification of peyote as a habit-forming and injurious drug. The subject concerns Indians in a dozen or more states; and it is hoped that the communication in *SCIENCE* will be given attention.

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Measuring Rate of Growth of Leucocytes

RECENTLY, Osgood, Li, Tivey, Duerst, and Seaman (1) reported a method for quantitatively measuring the rate of growth of leucocytes in tissue culture, by determining DNAP³² (radioactive phosphorus in deoxyribonucleic acid). In the opinion of the writer, the paper presents no acceptable evidence for the validity of the method. On the contrary, it serves to illustrate a number of errors that must be avoided in this type of work.

Interest in DNAP³² has been stimulated by the fact that its rate of formation is so low that it may be dependent upon the rate of formation of new DNA, associated with mitosis. Indeed, advantage has been taken of this fact (2) to obtain a qualitative indication of mitotic activity. But, as Hevesi has remarked (3), it is of the utmost importance to purify the sample of DNA very carefully, in order to remove phosphorus compounds that may have a much higher specific activity. Many workers (3-5) have perfected elaborate methods for isolation of DNA and have presented analyses to characterize their preparations—in other words, to show that DNA was actually isolated and that DNAP³² was actually determined.

For isolating DNA, Osgood *et al.* utilized a procedure introduced by Schmidt and Thannhauser (6)—alkaline hydrolysis of ribonucleic acid under conditions that do not render DNA acid-soluble. The authors of the Schmidt-Thannhauser method did not specifically study its application to blood leucocytes, nor did they claim that it offered the precision necessary for isolation of DNAP³² of any tissue. Furthermore, Osgood *et al.* have omitted the important first part of the method, without any stated reason. Thus their method for determination of DNAP³² was essentially a new one, but they presented no analytical data of any sort to justify calling their preparation "DNA." Nevertheless, it is possible to judge the soundness of their procedure, from certain papers that they do not cite. (These studies were concerned

with tissues other than blood leucocytes. Although Davidson, Leslie, and White (7) applied the Schmidt-Thannhauser method to bone marrow, they apparently did not confirm their results by use of any other method for determining nucleic acids.)

Schneider (8) prepared DNA from six different rat organs, according to Schmidt and Thannhauser. To this "DNA" he applied his hot trichloroacetic acid extraction procedure, which, as he has shown, separates nucleic acid from non-nucleic acid phosphorus. It was then evident that Schmidt-Thannhauser "DNA phosphorus" was 3-66% too high (compare columns IV-D and V-B of his Table I). Later the Schmidt-Thannhauser method, modified as suggested by Schneider, was applied by the present writer with others (2) to DNAP³² determination in whole rat liver and hepatomas. It was also applied to nuclei isolated from these tissues. "DNAP³²" values were greatly reduced, in most cases, if cytoplasm (containing no DNA) was removed. Thus the Schmidt-Thannhauser procedure is not generally applicable to determination of DNAP³² in whole tissues. Whether Osgood *et al.* actually isolated DNAP³², cannot therefore, be determined from their preliminary report.

That the true rate of DNAP³² formation in the cultured leucocytes was actually much lower than the reported rate is suggested by the observations disclosed concerning the degree of vitality possessed by the cells. Generally speaking, the cell counts graphically presented in Figs. 1 and 2 take the form of logarithmic death curves. It must be remembered that it is not possible, with Osgood's techniques, to separate the mitotically active cells, if such are present, from those that are dying. In a paper on marrow cultures (9), which Osgood *et al.* cite, Osgood states that his cultures "show degeneration after one to three weeks, although mitoses have been found at thirty-four days. . ." However, in the tracer experiments reported recently, blood leucocytes, not marrow cells, were cultured, and the culture conditions were stated to be especially unfavorable. Apparently, no mitotic figures were observed.

Finally, Osgood *et al.* claim that by determining DNAP³² it is possible "to obtain a quantitative measure of the rate of formation" of new cells. As has been remarked (5), the immediate precursor or precursors of DNA phosphorus are unknown. Until they can be identified and the specific activity of phosphorus entering DNA can be determined, there can be no quantitative estimate of the rate of formation of DNA, or of new cells, by such an approach.

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IN ANSWER to Cunningham's criticism, we agree that DNA used for specific activity determinations must be carefully purified to remove labeled P^{32} of other compounds of higher specific activity which may be adherent to the precipitated DNA. That this is accomplished by washing the precipitated DNA 6 times with 0.5 N HCl in 5% trichloroacetic acid is shown in our article by (a) the data in Table 1, in which precipitated DNA from brain, skeletal muscle, and cartilage contained readily measurable DNAP³¹ and non-DNAP³¹ and P^{32} , but no detectable DNAP³²; and (b) by the failure to find DNAP³² in precipitated DNA from 37° C cultures of leucocytes from patients with chronic lymphocytic leukemia, or in control cultures of granulocytic leukemias kept at 4° C, despite the presence of labeled non-DNAP³² in the same cells.

The citation of methods for isolation and purification of DNA used by others (1-3) is not pertinent to the criticism of this paper; the method selected for our purposes was the simplest and most satisfactory tried.

The criticism of the failure of Schmidt and Thannhauser to apply their method to leucocytes is irrelevant. Most physicians would agree that a method that they showed to be applicable to thymus tissue would also be suitable for trial on histologically similar leucocytes.

The first part of the Schmidt-Thannhauser (4) method was omitted, after parallel analyses on human spleen (from a P^{32} -treated leukemic patient) by the original and modified Schmidt-Thannhauser techniques yielded comparable DNAP³¹ values and specific activities on the precipitated material identical with the chemical and radioactivity assays within the limits of error. The omission was made to simplify the procedure, since only total and DNA phosphorus specific activities were necessary for this paper. Our analyses prior and subsequent to those of the criticized article by the original and modified methods suggest that there is a loss of phosphorus measured as DNA in the extensive pretreatment procedure, but specific activities remain unaltered. The modified method was further checked by quantitative recovery of DNAP³¹ from nucleoprotein (prepared by the method of Mirsky and Pollister [5]) that was added to solutions containing serum, albumin, erythrocytes, or even egg yolk. Thus this simple modification, proved not to affect DNA specific activity, does not constitute a "new method." These results are to be published elsewhere.

Cunningham is correct in stating that we present no analytical data to prove our preparation DNA. We followed the custom observed by Cunningham, in his paper quoted above and his references (5), (6), and (8), in letting the chemical procedure describe the product. In the course of these investigations, DNA/

10^{12} leucocytes was determined for 11 patients with granulocytic leukemia, and 15 patients with lymphocytic leukemia (48 and 52 determinations, respectively). These values were found to be normally distributed within the same distribution, with a mean value of approximately 600 mg/ 10^{12} cells ($S \sim 60$). Occasional determinations showed sperm DNA to be about one half this value. These data are not statistically different from the report of Davidson, Leslie, and White (6) for DNAP of human leukemic leucocytes of 699 mg/ 10^{12} cells (S.E. 194).

Cunningham's reference to the earlier paper of Davidson, Leslie, and White (7) seems irrelevant, unless he wishes to take exception to their work also. Their determinations were also based on precipitated DNA, and not calculated by difference as originally specified by Schmidt and Thannhauser.

Cunningham's reference to Table I in the paper by Schneider (8) shows only that DNA values calculated "by difference" (Column IV-D) are higher than those obtained directly upon the precipitated nucleic acid. This observation was also made in the preliminary studies done in our laboratory, and for that reason all calculations, as stated in our paper, are based on precipitated DNAP, not DNAP calculated "by difference."

Our critic's reference to his own work (9) on rat liver DNA seems scarcely comparable to DNA determinations on human leucocytes when one considers (a) the difference in species; (b) the difference in tissue composition—i.e., liver has not only liver cord cells, but ducts, vessels, and capsule as well; and (c) the apparently greater cytoplasmic volume of the rat liver cell in comparison with that of the human leucocyte. His observation that DNAP³² values of isolated nuclei were "greatly reduced" over those of whole liver cells is open to interpretations other than the one he gives. Inspection of the bottom rows of his Tables 1 and 2 (DNAP³²/P) will show the following: (Comparison of specific activities, not P^{32} alone, is here essential.) (a) the greatest decrease in DNAP³²/P is noted in his "control and 3'-MeDAB" values; (b) these "control and 3'-MeDAB" values are associated with the lowest DNAP³² values; (c) the "tumor" groups are associated with the highest P^{32} values; (d) the DNAP³²/P values for these "tumor" groups are for isolated nuclei, 111%, 73% and 102% of the whole liver values at 3 hr, 6 hr, and 6 days, respectively. Unless special precautions (not mentioned) are taken to remove statistical uncertainty of radioactivity determinations associated with low total P^{32} values, one would tend to be much more confident that the relative specific activities of the "tumor" group, which do not appreciably decrease, are much more reliable than those of the low-activity "control" or "3'-MeDAB" groups. His values in Table 2 are modified by his correction for loss of nuclei in separation: ("Judging from desoxyribonucleic acid determinations, 70-80 per cent of the nuclear material of the various whole tissues was recovered in the isolated nuclei, and the values shown in Table 2 were corrected

accordingly.") In view of these factors, and because of the failure to give standard errors, individual data, or other methods of assessing the confidence limits of either Table 1 or Table 2, it is difficult to evaluate the significance of his expression "greatly reduced." These data scarcely provide evidence for the sweeping assertion "Thus the Schmidt-Thannhauser procedure is not generally applicable to determination of DNAP³² in whole tissues."

It is true that peripheral blood was cultured in our experiments, but, as any physician knows, there are immature cells capable of division in the peripheral blood, identical with those in the marrow, of patients with leukemic leukemias. This fact is readily apparent from inspection of Figs. 1 and 2 of our paper. The data shown are based on 500-cell differential counts; when necessary, additional cells were counted to enumerate at least 50 cells "capable of division." Segmented neutrophils were present at 11 days in the cultures illustrated, whereas the average time of disappearance of segmented neutrophils from cultures of *normal* blood was 60 hr (10). This is further evidence that cell division and differentiation were occurring in these cultures of *leukemic* blood. Mitoses constituted about 0.5% of cells capable of division and were present in all smears of granulocytic leukemia cultures, but no statistically unassailable quantitative estimation was made of the mitotic index. No mitoses were observed in the cultures of cells from chronic lymphocytic leukemias. That cultures of cells from leukemic blood by our methods show mitoses has been confirmed by Gunz (11) and many others (12).

It is true that cell counts decrease over the period cultured. Particular attention is invited to the fact that DNAP³² uptake drops off as the "cells capable of division" fall below 100/mm³ for granulocytes, or 400/mm³ for lymphocytes (Figs. 1 and 2). No apologies are made for suboptimal cultural conditions; these were sacrificed to insure enough cells for analysis of comparable volumes of cells in patient and culture, capable of yielding unequivocal chemical analyses, without the use of carrier compounds. The presence of dead and disintegrated cells in these cultures leads to underestimation, not overestimation, of the relative rates of DNA-uptake of P³².

It is quite true that the immediate precursors of DNA are unknown, but it is obvious that inorganic radioactive phosphorus introduced into culture medium or the patient's plasma is the primary precursor of labeled DNAP³². The use of total medium or plasma phosphorus specific activity as an interim quantitative guide would make our estimates err on the conservative side. The phenomenon of saturation of DNAP³² specific activity equivalent to the specific activity of the patient's plasma, seen in Fig. 1 (and seen repeatedly in other patients with chronic granulocytic leukemia [13]), strongly sug-

gests complete turnover of DNAP³², especially as later studies have shown that the maximum specific activities of leucocyte acid-soluble and lipid fractions are comparable to these DNA saturation values.

Careful reading of our article will disclose that "quantitative" claims are made only in the sense of relative rates between the *in vitro* and *in vivo* human experiments, and for differences in uptake rates between cells of the lymphocytic series from patients with acute and chronic leukemias, and between cells of the granulocytic series and cells of the lymphocytic series. A complete quotation of our original sentence by Cunningham would have made this clear to the reader of his criticism. The desirability of reducing this type of data to a series of chemical steps, the reaction rates of which could be predicted by appropriate differential equations, is not questioned, but both limitations of chemical knowledge and biological variation prevent us (and Cunningham) from such accomplishments at present. Such limitations, however, do not prevent Cunningham *et al.* (9) from stating at the conclusion of their cited article, in which mitotic rates are also not given, "Therefore the degree of incorporation of the isotope in deoxyribonucleic acid could be correlated roughly with the degree of mitotic activity."

In conclusion, may we say that the purpose of our paper in SCIENCE was to present conclusive evidence that growth was occurring in cultures containing up to 10 g of human leucocytes, to describe simple modifications of existing methods, and to present further evidence that DNAP³² uptake is a measure of the relative rates of new cell formation. The criticisms raised appear to us to originate in misinterpretation of the original article and in debatable interpretation of references, and they in no way invalidate the statements in the original article.

EDWIN E. OSGOOD
HAROLD TIVEY

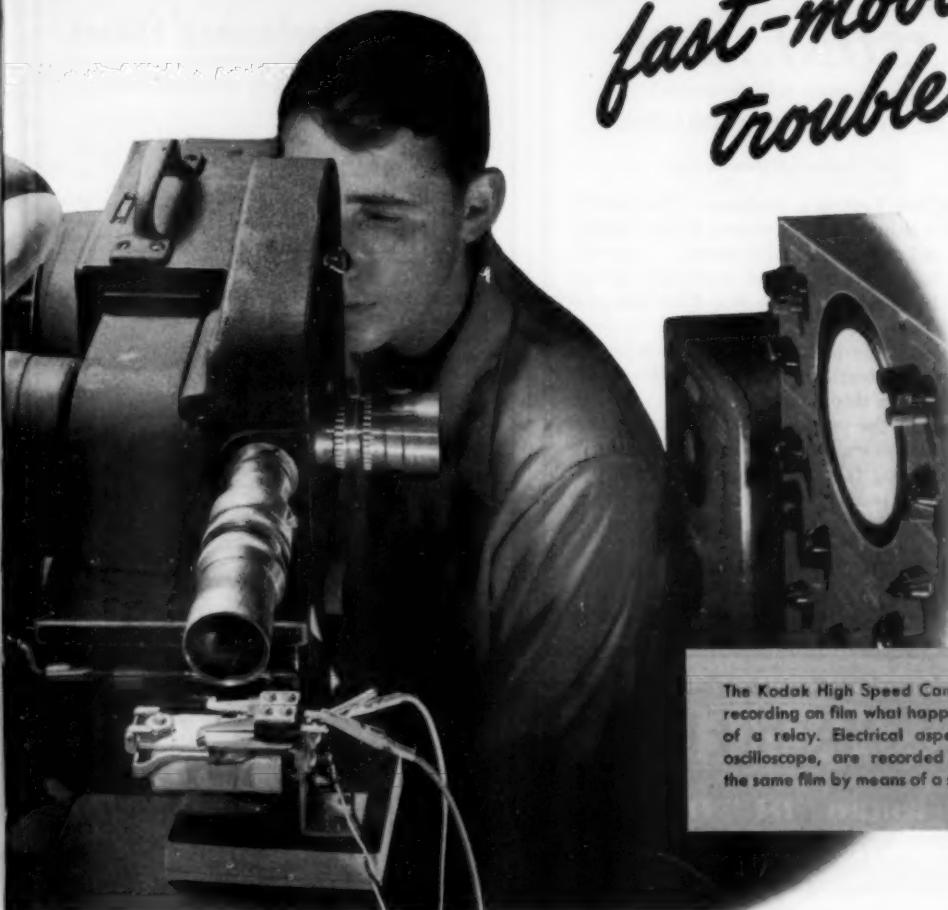
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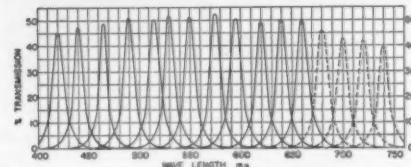
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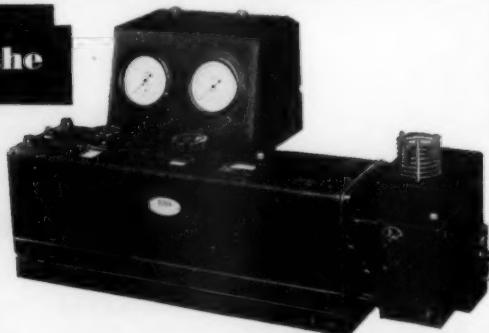
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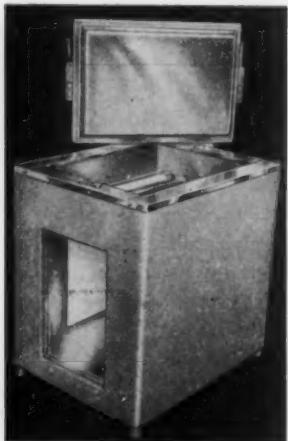
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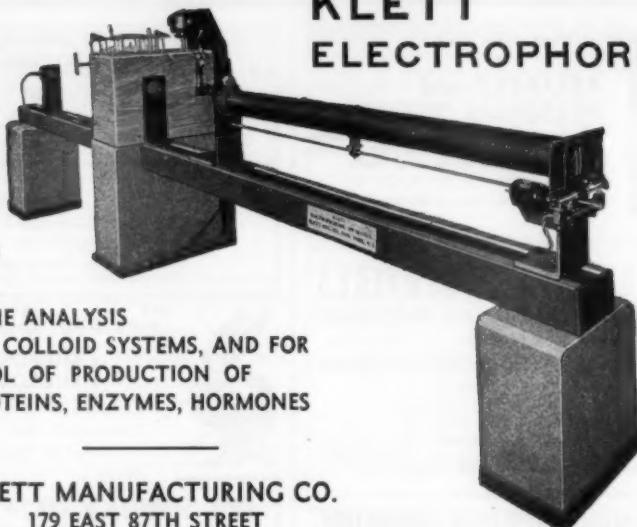
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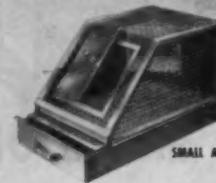
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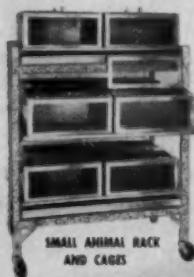
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ILLUSTRATED



SMALL ANIMAL RACK
AND CAGES



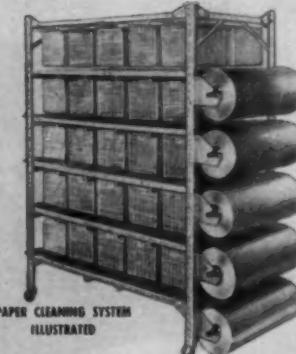
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CAGES AND RACK



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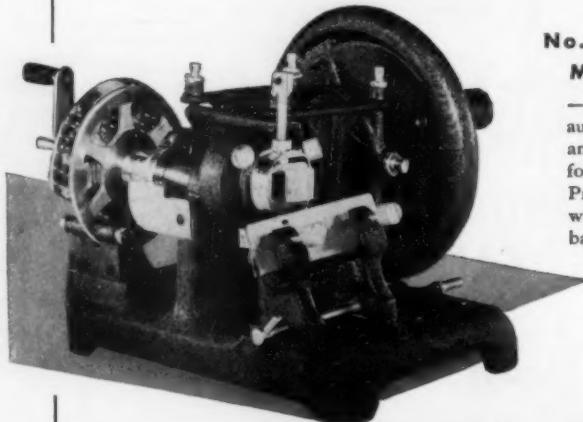
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